ANNUAL REPORT OF PROGRAM ACTIVITIES

NATIONAL INSTITUTE OF NEUROLOGICAL DISEASES AND STROKE

FISCAL YEAR 1973

PART II

U. S. DEPARTMENT OF HEALTH, EDUCATION, AND WELFARE
PUBLIC HEALTH SERVICE · NATIONAL INSTITUTES OF HEALTH

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U.S. DEPARTMENT OF HEALTH, EDUCATION, AND WELFARE
Public Health Service National Institutes of Health

ANNUAL REPORT
July 1, 1972 through July 30, 1973
Associate Director, Collaborative and Field Research
National Institute of Neurological Diseases and Stroke
National Institutes of Health

The goal of the Collaborative and Field Research (C&FR) program of NINDS is to conduct directed research of importance and relevance to neurologic diseases and communicative disorders. The work conducted in this program is a combination of in-house and out-of-house efforts which are chiefly by means of contracts. A number of the studies underway are collaborative studies which encompass the input of a number of outside institutions.

The C&FR program underwent extensive reorganization last year. There was some modification and elaboration of this reorganization during the current fiscal year and additional reorganization is required. For example, a new section was established-the Section on Biomedical Engineering and Instrumentation. The head of this new section is Dr. George Murray, formerly the head of the Section on Communicative Disorders. We are fortunate to have a man who has excellent capability in this area to organize our program. This new section was organized because there is a need for a central group to carry on development of neurologically useful diagnostic and therapeutic equipment through the prototype and clinical evaluation stages. This section will eventually require additional personnel. In the meantime, the work will be carried on by means of contract with a small in-house effort.

The transfer of Dr. Murray to the Section on Biomedical Engineering and Instrumentation leaves a vacancy for the position as head of the Communicative Disorders Section, and we are currently recruiting and plan to hire an individual for this position early in the next fiscal year.

The Infectious Diseases Branch, which was organized during the previous fiscal year, was further organized during this fiscal year. The Branch was functionally divided into three sections as follows: Immunochemistry and Clinical Investigations, Virology and Bacteriology, and Experimental Pathology.

The position of the Perinatal Research Branch was further solidified during this year by making Dr. J. S. Drage the Branch Chief. He had been Acting Branch Chief for approximately 11 months. In addition, the Section on Pathology, which consisted of a pathologist and five technicians was abolished and all of the work on neuropathology and general pathology and placentology for the Perinatal Project was placed on contract.

During the last fiscal year, the Laboratory of Central Nervous System Studies, headed by Dr. Carleton Gajdusek was transferred from C&FR to the Intramural Program of NINDS. This was done at the request of the Associate Director, C&FR, who after an assessment of this program came to the conclusion that it was intramural in nature.

By the end of June 30, 1973, there will be a total of 87 active contracts in NINDS. These contracts will be divided as follows: C&FR - 73, IR - 9, and EP -5. The generation of this number of contracts requires a great deal of work such as initiation of requests, advertisement, review of proposals, negotiations, technical monitorship, keeping of records, reception review, storage and dissemination of reports. It also requires a uniform method for all of those functions. To date, most of this work has been carried by the administrative officers of C&FR in addition to their other duties. The heavy work load produced by this contract program has necessitated the formation of a special group. Currently we are planning the formation of a contract operations group or office which will function under the direction of the Associate Director. We plan to have this office functional early in the next fiscal year.

Certain key personnel additions were accomplished this year to strengthen the various programs either underway or planned. Dr. G. F. Molinari, a neurologist with excellent background in the field of head injury and stroke, was added to the staff of C&FR. He is head of the Section on Head Injury and Stroke in the Applied Neurologic Research Branch and is now in the process of organizing a research program for this important area.

In addition, Dr. Karin Nelson, also a neurologist, was added to the staff of the Perinatal Branch. She is much needed in data analysis relating to neurological diseases of the Perinatal Project.

Recently, Dr. W. Watson Alberts joined the staff of C&FR as a Special Assistant to the Associate Director.

Two of the branches of C&FR were transferred from Building 36 on the Campus to the Federal Building. They were the Epidemiology and Special Programs Branches.

This last fiscal year was an especially arduous year for the top staff of the C&FR Program; first, because of the advent of a new Associate Director; and second, because of the reduction in funding during this fiscal year and the potential further fund reduction in the upcoming fiscal year. Consequently, two major efforts were initiated. One was an indepth review of all ongoing programs, and second was the initiation of integrated program planning.

A thorough review was made of all programs underway from the standpoint of scientific merit, relevance, and priority. These reviews were a combination of inhouse reviews by the Associate Director and also reviews with the help of separate advisory committees consisting of outside experts. The results of these reviews will be reflected in the FY 74 and FY 75 programs. As a result certain programs will be enhanced, others will be reduced in scope or dropped, and in certain cases new programs will be initiated.

An example of reviews undertaken was a thorough evaluation of the Special Programs Branch which is responsible for the Neurological Information Network. This program which was started as an experiment in 1966 for

consolidating and disseminating research information to the outside community was extensively reviewed by the Scientific Information Program Advisory Committee during the past year. The Associate Director of C&FR held three separate meetings with this group during which they addressed themselves to such questions as should NINDS support such an activity? If so, what kind of information program would be the most effective and most useful to conduct for the benefit of the neurological and communicative disorders community? A report of the findings of this survey will be presented to the Director of NINDS in the near future.

The Epidemiology and Infectious Diseases Branches have not had their programs thoroughly reviewed by outside peer review groups in the past . A peer review group was organized recently composed of eminent bacteriologists, virologists, epidemiologists, and neurologists. It held its first two-day meeting in March 1973 and will hold another two-day meeting in July 1973. It will advise the Associate Director on the relevance, priority and scientific quality of ongoing programs and also will make recommendations for future required research. The Communicative Disorders program has been thoroughly reviewed in conjunction with its advisory committee and consequently a good program has been initiated. The Epilepsy Advisory Committee met twice in the last fiscal year and has reviewed the epilepsy program. The Perinatal Project is practically under constant review as its advisory committee, the Coordinating Committee for Data Analysis has been meeting with perinatal personnel on the average of once a month during the last year. This committee is being enlarged by the addition of a neurologist and a psychologist. The Section on Biomedical Engineering and Instrumentation which was initiated in January 1973 needs some outside advisory people to examine its ongoing program and help develop a more comprehensive program. This will be accomplished in the near future.

The C&FR program needs an overall advisory committee to examine its entire program from the standpoint of need and balance. We are planning to organize one soon. It will be composed of individuals with specialties in the various fields covered by the C&FR program. The individual specialists in conjunction with additional required consultants will be responsible for examining the various individual pieces of the overall program.

When the new Associate Director of C&FR arrived on the scene, he was told by the Director of NINDS that there was a need for programs of specific neurological diseases and communicative disorders. As a result, program planning was initiated to attempt to establish plans for programs in various disease areas. It was also clear that C&FR had a variety of experts available who were functioning exclusively in their own circumscribed specialties. By pooling these various talents in the eight branches and sections, it appeared possible to perform integrated program planning for various disease areas. This would be accomplished by organizing program planning teams for various diseases. At first, the plan was to use only C&FR personnel on these teams, but eventually the membership was broadened by including appropriate personnel from both the Intramural and Extramural programs of NINDS.

By placing a horizontal team organization across the conventional vertical organization of the Institute, C&FR thus created an all-inclusive grid to utilize the Institute's resources most effectively.

First, a tentative system was worked up to establish priorities for various neurological diseases. Stroke headed the list, other priority diseases were head and spinal injury, epilepsy, communicative disorders, multiple sclerosis and etc. At the time of the initiation of this program, we had no expert to lead either the stroke or head injury programs which would have been the logical diseases with which to start. Therefore, we decided to start with diseases in which we had some expertise and also some ongoing programs. We selected four disease areas for program teams: multiple sclerosis, communicative disorders, epilepsy and stroke and head injury. To date, we have actually initiated two program planning teams - Multiple Sclerosis and Communicative Disorders. We plan to start the other two soon. The functions of these planning teams is to (1) review ongoing work both inhouse and out-of-house, (2) determine the status of knowledge, (3) recommend an integrated program consisting of basic research (inhouse and grants); and directed research; and a budget required to carry on this work. After these programs have been reviewed internally, they will be exposed to groups of outside experts for their critical evaluation. Subsequently, they will be presented to the Director of the Institute for his approval prior to initiation.
In addition to program planning, various pieces of the program after initiation would be the responsibility of the various members of the teams in their areas of expertise. The chairmen of the team would be responsible for the overall status of this particular program. The Multiple Sclerosis Program is nearing completion and will be presented in the near future. The membership of the Multiple Sclerosis Planning Team is:

Dr. John Sever - Infectious Diseases Branch, C&FR (Chairman)
Dr. Edgar Bering - Special Programs Branch, C&FR
Dr. Roscoe Brady - Laboratory of Neurochemistry, Intramural Research
Dr. Jacob Brody - Epidemiology Branch, C&FR
Dr. Jonas Ellenberg - Office of Biometry, C&FR
Dr. Joseph Gibbs - Laboratory of Central Nervous System, Intramural Research
Dr. George Murray - Biomedical Engineering and Instrumentation, C&FR
Dr. O. Malcolm Ray - Extramural Programs
Dr. Henry D. Webster - Laboratory of Neuropathology, Intramural Research

We believe this type of planning will be the most effective utilization of funds and personnel for the Institute.

It was not possible to present the C&FR program for FY 74 in the format described above because of insufficient time. Therefore, each branch presented its program for FY 74 and the ensuing five years in an individual branch format. Each branch was required to list its goals, objectives, projects, milestones, and budgets for the next five years. Each branch was also required to arrange its program in two ways - a core program and optimum program. These will be presented to the Director of the Institute prior to the beginning of the next fiscal year for his review and approval.

Our other important matter which should be mentioned at this point is the completion and approval of the Comprehensive Plan for Analysis and Interpretation of Collaborative Perinatal Project Data. Serious work began on this plan last August. The plan was approved on April 20, 1973, and it is now being implemented.

At the direction of the Director of the Institute and under the Chairmanship of the Deputy Director of NINDS, the Associate Director was heavily involved in planning for a reorganization of the Institute. A draft plan was devised as a result of this effort.

APPLIED NEUROLOGIC RESEARCH BRANCH

This Branch which consists of two sections, the Section on Epilepsy and the Section on Head Injury and Stroke, is headed by Dr. J. Kiffin Penry.

Section on Epilepsy

At present, there is no head of this section and its work is being directed by Dr. Penry. We are now actively recruiting for a head of this section and plan to hire an individual early in the next fiscal year.

The epilepsy program has been chiefly devoted to studies of anticonvulsant drugs. This has involved a number of collaborative studies which are continuing. One of the newer studies which has been initiated is an important pilot study, conducted by the University of Kansas Medical Center, to determine the effectiveness of drugs in preventing post traumatic epilepsy. In other studies sponsored by the Branch, carbamazepine has been shown to be an effective anticonvulsant. It is hoped that during the next year the FDA will approve this investigational drug as the first new anticonvulsant to be approved in 14 years.

The Section continues to evaluate reliable methods for measuring minute levels of anti-epileptic drugs in serum, such as gas-liquid chromatography and radioimmunoassay.

In midsummer an epilepsy bibliography (1900-1950) will be published. Two important books were published during the year: Anti-Epileptic Drugs, and Experimental Models of Epilepsy-A Manual for the Laboratory Worker. An NINDS bibliography on Blood Level Determinations on Anti-Epileptic Drugs-Clinical Value and Methods was also published, and a motion picture, The Absence Seizure, was produced.

The epilepsy program will be enhanced during the next fiscal year by conducting studies on the feasibility of comprehensive epilepsy centers and possibly epidemiological studies. There are plans to initiate an Epilepsy Program Team.

Dr. Donald Bennett of the University of Utah will come on board as a visiting scientist for six months beginning on June 15, 1973. This program was materially aided during the last year by the fine efforts of the Epilepsy Advisory Committee.

Section on Head Injury and Stroke

Dr. G. Molinari was appointed Head of the Section during FY 73. He is now in the process of forming a program on head injury and stroke. He is currently organizing a planning team for this endeavor.

Although we had no leader for this Section until recently, a program in stroke had been initiated. This study involves a most important aspect of stroke, namely, Transient Ischemic Attacks (TIA). Neither the etiology, the natural history, nor the ideal treatment of TIA have been established. In order to gather data on this phenomenon, a cooperative study of hospital frequency and character of transient ischemic attacks was set up at six universities. This program has been in effect about eight months. A first review of this program produced a number of interesting findings. This program will run for another eighteen months.

The pilot phase of the Collaborative Study on Cerebral Survival has been completed, and we anticipate publication of the results during FY 74.

PERINATAL RESEARCH BRANCH

Dr. Joseph S. Drage was appointed Chief of this Branch during FY 73. This year marked the completion of the follow-up examinations of children at seven years of age who were born to 50,000 women at 12 collaborating U.S. Medical Centers. The data of children now eight years of age will be completed in 1974. As data collection draws to a close, major emphasis is being placed on data analysis and the preparation of reports for publication. To provide a framework and time schedule for this effort, a Comprehensive Plan for Data Analysis and Interpretation has been developed and approved. Implementation of the plan will be through teams of researchers in ten different areas. Each team will be headed by a member of the Professional Consultants Section of Perinatal Research Branch and on each team will be a statistician of the Office of Biometry Staff who will participate fully in the development of analysis. By the end of FY 76, monographs are to be completed in each of the following ten areas: (1) Cerebral Palsy, (2) Mental Retardation, (3) Communicative Disorders, (4) Visual Abnormality, (5) Convulsive Disorders, (6) Learning Disorders, (7) Minimal Brain Dysfunction, (8) Congenital Malformations, (9) Birthweight-Gestational Age Relationships (Prematurity), (10) Neuropathology, and General Pathology and Placentology.

The Coordinating Committee for Data Analysis has been very active during the last year. They have met with NINDS personnel on the average of once a month. Their input to the comprehensive plan and other phases of the perinatal project has been invaluable. This committee is being increased by the addition of a neurologist and psychologist.

The Section on Pathology was closed during this last fiscal year and all personnel reassigned. The work of this section has been placed under contract.

SECTION ON COMMUNICATIVE DISORDERS

This section was initiated last year under the direction of Dr. George Murray. Dr. Murray with the help of the Communicative Disorders Advisory Committee and the help of the internal Communicative Disorders Planning Team has

started an excellent program. Dr. Murray is currently Acting Head of this Section, and we are currently recruiting for a new head who will be hired early in the next fiscal year. There are also plans to hire an expert on speech for this section who will implement programs in this important area of research.

The Section on Communicative Disorders develops and carries out projects directed toward the diagnosis and treatment of disorders of hearing, speech, and language. Of particular concern are the diagnosis and remedy of infant and early childhood hearing impairment. Projects involving sensory aids to improve the lipreading ability of the deaf adult, and studies of the underlying requirements for precise tailoring of hearing aid characteristics are also underway. The fundamentals of speech and its rehabilitation are under study. The section concentrates on those projects not supported under the basic research grant mechanism and carries prototype development of instrumentation through the stage of clinical testing.

SECTION ON BIOMEDICAL ENGINEERING AND INSTRUMENTATION

This newly formed section, under the leadership of Dr. George Murray, is charged with overall coordination of the development of neurologically useful diagnostic and therapeutic technical equipment through the prototype and clincial evaluation state. In-house laboratory research and development is combined with collaborative projects undertaken by contract in laboratories of other institutions. The section serves to complement other facilities of NIH by directing its attention specifically to neurological disorders.

Some of the activities of the section include the non-invasive assessment of the status of intracranial tissue and the development of EEG research techniques. Methods of measuring regional cerebral blood flow will be investigated during the coming year.

This section needs an outside advisory committee which will be formed in the near future. This section will also require additional personnel.

INFECTIOUS DISEASES BRANCH

This branch, which was organized as a separate entity last year, is under the direction of Dr. John Sever. This year the branch was organized into three sections: Immunochemistry and Clinical Investigations, Virology, and Bacteriology, and Experimental Pathology.

The goal of this branch is to carry out planned directed research programs concerned with infections which damage the human nervous system. The research activities are divided into three areas: Perinatal, Acute, and Chronic.

The perinatal clinical research is further divided into (1) studies relating to the Collaborative Perinatal Project, and (2) cooperative and other studies.

The total perinatal area accounts for more than 65 percent of the research effort, acute infections about 15 percent of the effort, and chronic infections 20 percent.

Major reports are being developed for the Collaborative Perinatal Research Program. New experimental findings with infections in monkeys are providing tools for the study of infections and malformations. For example, experimental studies have clearly demonstrated the production of hydrocephalus in the fetus by the innoculation of pregnant monkeys with Flu-A or Venezuelan Equine Encephalitis virus. Cataracts are also consistently produced in the fetus with VEE.

Pregnant women, children, and cancer patients have been found to have been infected with cytomegalous virus for long periods. Leukemia patients have high rates of infection and this may have contributed to some of the clinical findings in this disease. In a collaborative program with Georgetown Medical Center, highly specific and sensitive new tests for cellular immunity to virus infections (e.g. measles and German measles) have been developed. It is planned to develop these tests for bedside use.

Dr. Sever, as Chairman of the Multiple Sclerosis Planning Team, has been heavily involved in this planning process.

This branch is under review by an outside peer review group for relevancy, scientific merit, and priority of its program. The review group met for two days in March 1973 and will hold its next meeting in July 1973.

EPIDEMIOLOGY BRANCH

This branch is headed by Dr. Jacob Brody. Its goals are to identify patterns of neurologic diseases in human populations and to define interactions of affected persons, etiologic agents and environmental setting.

The current projects are related to epidemiology, genetics, and the Guam studies of foci of neurologic diseases.

This branch has been heavily involved in the epidemiologic studies of amytrophic lateral sclerosis, multiple sclerosis, and subacute sclerosing panencephalitis. In the last year, the branch has moved toward examining diseases of higher priority such as stroke and epilepsy. A stroke epidemiologic study was recently initiated, through a contract with the School of Public Health at the University of Texas. They are studying the stroke risk factors in selected black populations to determine the relative role of environment and heredity in the observed higher rates of stroke among American blacks. Through a contract with the Kaiser Permanente Foundation in San Francisco, the branch is studying a population which developed strokes at some time after a multiphasic screening examination. In collaboration with the Epidemiology Branch, NHLI, the branch is studying patterns of stroke among resident and migrant populations in California.

Last year a peer review group met in Guam to review the Amyotrophic Lateral Sclerosis and Parkinson's Dementia program and make recommendations. This

group made certain recommendations for work to be finished within a year. They indicated that it would be advisable to have a well qualified epidemiologist present at Guam for one year in order to see that the work was completed. Dr. Dwayne Reed, Deputy Branch Chief, was sent to Guam for a one year assignment on January 1, 1973. The peer review group for the Guam program will meet in Guam again in September 1973. At that time, we hope to have a recommendation as to whether to continue or discontinue this program.

The overall epidemiology program is being reviewed by the same peer review group which is reviewing the Infectious Diseases Program. As indicated previously, this group will meet again in July, 1973.

Dr. Brody has also been involved in the activity of the Multiple Sclerosis Program Team. During the current fiscal year, the Epidemiology Branch with the exception of its laboratory was moved from Building 36 to the Federal Building.

SPECIAL PROGRAMS BRANCH

The Special Programs Branch is headed by Dr. Edgar Bering. Its goals are first, to organize programs for the exchange of scientific information so that there will be a more rapid and efficient use of research results and, second, the development of NINDS related research in foreign countries through the PL-480 program.

The main task of this branch is to run the Neurological Information Network. This consists of the Brain Information Service (BIS) at UCLA, the Information Center for Hearing, Speech, and Disorders of Human Communication (ICHS & DHC) at Johns Hopkins University Medical School, the Clinical Neurological Information Center (CNIC) and the Cerebrovascular Alerting Service (CVI) at Mayo Foundation. The branch has continued to produce Parkinson's Disease and Related Disorders, an alerting bulletin of relevant citations which comes out monthly.

The Neurological Information Network which started out as an experiment in 1966 has grown considerably in size and expense since its inception. It was decided this year that this would be a propitious time for thorough review of the entire program. Certain basic questions were asked in this review such as: Is this the type of activity which NINDS should support? If so, what is the most efficient service which could be organized and maintained by the Institute for the benefit of the biomedical community? How effective has this information network been?

The Scientific Information Program's Advisory Committee met three times during this last fiscal year to evaluate the program and provide recommendations to the Associate Director, C&FR. A report will be made to the Director of the Institute in the near future.

During the year this branch was moved from Building 36 to the Federal Building.

Dr. Bering has been heavily involved in the Multiple Sclerosis Program Team.

OFFICE OF BIOMETRY

Mr. William Weiss is the chief of this branch. The Office of Biometry is a service organization which provides support to all sections of the Institute as follows: statistical design and analysis, computer science and systems analysis.

The Office of Biometry is very much involved in the implementation of the Comprehensive Data Analysis Plan of the Perinatal Project. Six of their personnel have been assigned to specific areas mentioned in the comprehensive plan of the Perinatal Project. The Biometry people so assigned have the responsibility of working very closely with their counterparts from the Perinatal Branch and are involved from the very inception in each of the ten major project areas.

In addition to the Perinatal Project of C&FR, the Office of Biometry is also involved in C&FR projects as follows: Infectious Diseases, Epidemiology, and Applied Neurologic Research. The Office of Biometry is also participating in projects with a number of laboratories and branches of the Intramural Program plus some work for the Extramural Program.

The Office of Biometry made a major contribution toward the planning of the NINDS Reorganization Plan.

The branch chief of the Office of Biometry is a member of the Coordinating Committee for Data Analysis for the Perinatal Project.

NEURAL PROSTHESIS PROGRAM

In the past the Sensory Prosthesis Program was limited to feasibility studies for the development of a visual prosthesis for the blind through the use of direct cortical stimulation. During the past year the program, which is supervised by the Laboratory of Neural Control of the Intramural Program, broadened its scope to become the Neural Prosthesis Program by initiating projects in electrical stimulation to initiate emptying of the bladder and in neuromuscular stimulation for the purpose of producing functional contraction of voluntary muscle. During FY 75 it is planned to add two C&FR positions to this program.

CONCLUSION

The foregoing is a summary of the work of C&FR during Fiscal Year 1973. As can be seen, program planning is becoming an increasingly important element. Wherever possible, there is an integration of the talents which exist in the various branches, especially relative to conducting research programs in specific disease areas. Program planning teams are being used as indicated. There is also an attempt to bring in expertise from the other major organizational elements of NINDS.

All C&FR programs are being reviewed for scientific merit, relevance, and priorities.

The contract function which is a large element of the work load of C&FR is being modified, improved, and organized.

ANNUAL REPORT
July 1, 1972 through June 30, 1973
Section on Communicative Disorders, C&FR
National Institute of Neurological
Diseases and Stroke
George C. Murray, Ph.D.

Introduction:

The Section on Communicative Disorders was established in the Office of the Associate Director for Collaborative and Field Research on April 12, 1972, to develop and carry out planned directed research toward the improvement of diagnosis and treatment of disorders of hearing, speech and language. In addition, the Section supports and coordinates the Communicative Disorders Planning Team and the Ad Hoc Advisory Committee on Communicative Disorders for C&FR, NINDS. The principal areas of activity currently include:

1. Hearing Health and Auditory Disorders

Projects have been initiated for studies aimed at the development of improved methods of detection and assessment of hearing impairments in young children. A contract directed study by Stanford Research Institute is developing and evaluating a three-stage screening and diagnosis program for the early detection of potential hearing impairments in infants and prelinguistic children.

Under an existing contract with the Information Center for Hearing, Speech and Disorders of Human Communication at Johns Hopkins University, a survey has been designed and pilot tested which will assess the current practices and attitudes both of the lay public and of the health professionals concerning the early detection of infant hearing impairments and to identify impediments to such detection.

Contracts have been solicited and are being negotiated for epidemiologic studies of distinctive patterns in the medical, social, economic, and familial factors of hearing impaired individuals; the aim being to identify causal and high risk factors to guide further studies in the specific etiologies of hearing losses.

2. Speech and Language Disorders

With the assistance of the Communicative Disorders Planning Team, and with the advice of the Ad Hoc Advisory Committee on Communicative Disorders, the Section is developing specific project plans for studies of aphasia, and the assessment of language capabilities and treatment of language disorders. Plans are also being developed for studies of comprehensive measures of speech impairments, fundamental elements of speech, and the treatment of speech disorders. However, no active research projects were initiated in these areas this year.

3. Aids to the Communicatively Impaired

Research and development projects begun this year are aimed at improving the effectiveness of the prescription, fitting, and use of aids to the communicatively impaired. A program was begun last year to study the relationships between the residual auditory capacities of a hearing impaired individual and the configuration of electroacoustic hearing aids which will optimally improve his speech communication capabilities. To instrument such studies, design concepts have been developed and prototypes are being provided through contracts with Bolt Beranek and Newman and CBS Laboratories for clinical research hearing aids which will provide, in a wearable form, the capabilities of a tailorable master hearing aid. Proposals to carry out a pilot study of the clinical evaluation of the effectiveness of various parameters of hearing aid characteristics have been solicited and are being negotiated.

A project begun last year under a contract with Johns Hopkins University has been studying the use of patterned tactile and visual cross-modality presentation of speech derived information as a feedback of deaf infant's vocalization and its effectiveness on the development of his vocalization patterns.

Contract proposals have been solicited to study methods for developing a speech analyzing aid to assist deaf individuals in lipreading by supplying supplemental cues derived from acoustic speech signals.

Work on aids to the handicapped is being done in collaboration with the Section on Biomedical Engineering and Instrumentation of C&FR, NINDS.

CONTRACT NARRATIVE
Communicative Disorders Section, C&FR, NINDS
Fiscal Year 1973

CBS LABORATORIES (NINDS-72-2316)

Title: Design Concepts and Electronic Configurations for Wearable Master Hearing Aid Device for Clinical Studies

Contractor's Project Director: Emil L. Torick

Current Annual Level of Support: $69,753

Objectives: The contractor will provide design concepts and develop and deliver prototype models of a wearable master hearing aid device for clinical research studies. The aid will be a two channel aid capable of monaural, straight binaural, or cross or bi-cross fitting, and whose response characteristics in terms of gain, slope of gain versus frequency, bandwidth, upper and lower roll off limits, maximum power output, form of compression, and attack and release times of compression can be independently varied by the investigator.

Major Findings: Successful design concepts and prototype instrument development will be completed and prototype instruments delivered to the NIH by the end of FY '73. Target specifications will be met with the possible exception of the inability to provide the full 6kHz bandwidth higher than 120dB out. The remaining high level of 120 to 140dB will be restricted to 4kHz if necessary.

Significance to NINDS Program and Biomedical Research: For the estimated 5 to 10 million Americans with hearing impairments of a handicapping degree the primary rehabilitative treatment measure is the fitting of appropriate electroacoustic hearing aids. Clinical studies of relationships between the individual residual hearing characteristics of the impaired subject and the characteristics of electroacoustic aids which optimally improve speech recognition capabilities can be carried out with the use of this clinical research hearing aid. The successful establishment of such relationships would allow more effective use of the recent developments both in diagnostic procedures and in electronic capabilities to better fit tailored hearing aids to the specific needs of hearing impaired individuals. Since speech communication is fundamental to everyday life for most individuals, and since interruption of such communication capabilities is basically a neurologic problem affecting so many Americans, the development of improved treatment through better hearing aids is of major importance to NINDS programs.

Cooperating Units: The technical supervision of the instrument development in this project is provided through cooperation with the Section on Biomedical Engineering and Instrumentation, C&FR, NINDS.

Proposed Course of Contract: This contract will be completed in June of 1973 and the final report and prototype instruments will be delivered to NINDS by July of 1973

CONTRACT NARRATIVE
Communicative Disorders Section, C&FR, NINDS
Fiscal Year 1973

STANFORD RESEARCH INSTITUTE (NINDS-72-2317)

Title: Design, Test, and Validate a Three-Phase Screening Program

Contractor's Project Director: E. James Kreul, Ph.D.

Current Annual Level of Support: $98,900

Objectives: The contractor will devise and evaluate methods for the early detection of hearing impairments among children below the age of 5 years. Procedures will be designed for use at three levels of increasing expertise by parents, by non-specialist professionals and by audiologists, respectively. The contractor will provide data and analysis of the reliability and validity, as well as complete instructions for conducting these screening procedures.

Major Findings: A variety of easily fabricated sound-generating devices have been made and analyzed. Instructions have been written and tested for use at levels one and two. Four subcontracts have been signed to enable a multifaceted attack. A high-risk registry has been initiated, and testing of children has begun. About one thousand tests will have been completed by the end of the year. Reliable acoustic reflex responses in neonates have been obtained at one of the subcontracting institutions. Recognition of sound-evoked responses by parents has been encouraging.

Significance to NINDS Program and Biomedical Research: Early detection of hearing impairment enables the application of remedial measures in the period critical to the development of communicative ability, so as to reduce the degree of communicative handicap known to result from deafness in childhood. The associations between deafness and causative factors are more amenable to investigation when the problem can be identified at its onset. The population under age 5 years has been largely neglected in this regard because of lack of adequate detection methods.

Proposed Course of Contract: The contract is expected to run for three years to enable longitudinal study of cases necessary to assess the incidence of false positives and false negatives as a consequence of the tactics employed.

CONTRACT NARRATIVE
Communicative Disorders Section, C&FR, NINDS
Fiscal Year 1973

JOHNS HOPKINS UNIVERSITY (NINDS-72-2318)

Title: Study of Cutaneous and of Visual Patterned Stimulation Communication Aids for the Profoundly Deaf Infant

Contractor's Project Director: Moise H. Goldstein, Ph.D.

Current Annual Level of Support: $25,000

Objectives: The contractor will develop techniques for and conduct comparative evaluation studies of the use of cutaneous and of visual dynamic patterned stimulation derived from deaf infant's vocalizations to provide for feedback for vocalization development and as a communication aid for profoundly deaf infants. Comparative studies are being made on three groups of profoundly deaf infants in the age range of 12 to 24 months to determine the effectiveness of visual patterned stimulation and tactile patterned stimulation on the development of vocalization in these infants during and following training sessions, compared to the development of a group of similar subjects receiving standard teaching techniques without the use of the prosthetic aid. The contractor will assess the feasibility of using such processed speech feedback through alternate sense modalities as a speech training and communication aid for the severely deaf infant, and establish whether cross modality transfer from one sense modality to another is indeed effective.

Major Findings: Instrumentation development has been completed and pilot tested on three normal and three severely impaired infants to develop the protocols and training procedures to be used in the study of additional groups of such infants. Preliminary indications show some effects of the use of the training aid but statistical significance of the relative effectiveness of the three training procedures must await the testing of additional patients in the coming year.

Significance to NINDS Program and Biomedical Research: It has long been felt that normal development of language patterns and speech skills require both participation in speech communication using normal language patterns and practice in the mechanical skills of articulation and vocalization during normal developmental periods. The severely deaf infant is denied such input of acoustic speech information both from others and from his own vocalization efforts by virtue of his loss of hearing. It is felt that providing for increased sensory input of the patterns of normal acoustic speech during the normal developmental period through cross modality input with such prosthetic aids, may assist in a more normal development of language and speech capabilities during the early years. If such cross modality sensory input proves effective, the later development of wearable communication aids based on such cross modality input could help the language and speech development of an estimated 250,000

pre-school children who suffer from hearing deficiencies sufficient to impair their normal language development.

Proposed Course of Contract: During the second year further tests will be made on an additional 18 patients and analysis and evaluation of the effectiveness of such training will be made to determine the feasibility of this approach to cross modality communication aids for profoundly deaf infants.

CONTRACT NARRATIVE
Communicative Disorders Section, C&FR, NINDS
Fiscal Year 1973

BOLT BERANEK AND NEWMAN (NINDS-72-2327)

Title: Development of a Wearable Master Hearing Aid

Contractor's Project Coordinator: Mr. Edward Starr

Current Annual Level of Support: $78,500

Objectives: The contractor will develop design concepts and provide three prototype models of a clinical research hearing aid which will perform as a wearable version of a master hearing aid. The characteristics of such an aid will be tailorable by the investigator in the dimensions of gain, slope of gain versus frequency, bandwidth, independent upper and lower cutoffs, maximum power output, compression, and attack and release time of compression. This aid will be binaural with two independent channels and also provide for cross and bi-cross connection.

Major Findings: Designs have been developed and instrumented which will accomplish the target specifications with the only notable exception that the wide bandwidth will be achieved only to the level of 120dB output and that in the 120 to 140dB range the bandwidth will necessarily be restricted. All of the other parameters can be successfully varied independently and the complete two cannel wearable aid can be packaged as a wearable device with head mounted microphones and earphones connected by a lightweight flexible cord to a shirt pocket or belt work electronics package. Prototype models for use in pilot studies will be available by the end of the fiscal year.

Significance to NINDS Program and Biomedical Research: It is estimated that between 5 and 10 million Americans have a partial hearing impairment of handicapping degree for which the primary rehabilitative and treatment measure is the fitting of an electroacoustic hearing aid. Recent advances in techniques and capabilities of audiometric assessment of hearing impairment and advances in the electronics capabilities of producing electroacoustic hearing aids of higher quality and more sophisticated tailored configurations should make it possible to provide these hearing impaired subjects with aids specifically tailored to their hearing needs. The basic knowledge of relationships between residual auditory capabilities of the hearing impaired individual and the aid characteristics which should be prescribed for him remain to be determined through clinical studies. Research hearing aids of the sort developed under this project are required for such clinical studies. Since functional disorders, which impair successful human communication via speech, are basically neurologic problems affecting a large segment of the population, the development of improved rehabilitation and treatment of these disorders is of major importance to the programs of NINDS.

Cooperating Units: This project is being carried out in cooperation with the Section on Biomedical Engineering and Instrumentation, C&FR, NINDS, who provides the technical direction of the instrument development.

Proposed Course of Contract: This contract will be completed in June 1973 and the final report and prototypes delivered by the end of July 1973.

ANNUAL REPORT
January 9, 1973 through June 30, 1973
Section on Biomedical Engineering and Instrumentation, C&FR
National Institute of Neurological
Diseases and Stroke
George C. Murray, Ph.D.

Introduction:

The Section on Biomedical Engineering and Instrumentation was established in the Office of the Associate Director, Collaborative and Field Research, NINDS on January 9, 1973. The Section carries out planned directed research to develop biomedical instrumentation needed to advance the diagnosis and treatment of neurological and sensory disorders. The Section develops and directs programs to appropriately adapt and apply recent advances in technology to problems in identified areas of neurosciences for which biomedical instrumentation is needed. The Section coordinates projects with other labs and branches of the Institute and the other biomedical engineering activities of the NIH. By working directly with the clinicians and scientists on their instrumentation problems, the Section seeks to provide the necessary interdisciplinary interface between the clinical problems and the technical development to assure the proper clinical input at all stages of development.

The laboratory research and feasibility studies of the Section are designed to explore possible approaches, adapt basic techniques to specific clinical problems, and to investigate the feasibilities of alternative methods.

The Section develops and directs prototype instrumentation engineering to adapt feasible approaches and techniques into practical clinical prototype instruments. The Section monitors the clinical testing and evaluation of prototype instrumentation and provides for needed modification and refinement to lead to useful practical clinical tools ready for commercial development and wide application.

Work in Progress:

The Section staff has in the past year assisted other branches and sections with their instrumentation development projects by providing technical advice, and as project officers provided technical supervision of several contracts. In addition, the Section has helped develop Requests for Proposals on new projects.

Gamma Camera Development:

As part of the program of the Section on Head Injury and Stroke, Applied Neurologic Research Branch, C&FR, a new semiconductor gamma camera for nuclear medicine is being developed under contract with Ohio State University (NIH-NINDS-72-2323). The technical aspects of this instrumentation development project are monitored and directed by the Head of the Section on Biomedical Engineering and Instrumentation. (See Annual

Report of ANRB, C&FR, NINDS).

Wearable Master Hearing Aids for Clinical Research:

The engineering development of clinical research hearing aids for use by the Section on Communicative Disorders, C&FR, are being developed under contracts directed by the staff of the Section on Biomedical Engineering and Instrumentation. Contract projects with Bolt Beranek and Newman, Inc. (NIH-NINDS-72-2327) and with CBS Laboratories (NIH-NINDS-72-2316) have developed designs and are providing prototype models of clinical research hearing aids whose characteristics are independently tailorable by the investigator in terms of bandwidth, upper and lower frequency cutoff, slope of gain frequency response curve, compression form, compression attack time and release time. Six prototype aids will be completed and delivered to NINDS before July 1973. (See also Annual Report of Section on Communicative Disorders, C&FR, NINDS).

Ambulatory EEG Recorder:

The Section on Biomedical Engineering and Instrumentation, C&FR, has collaborated with the Epilepsy Section, ANRB, C&FR and solicited proposals for a contract directed development of a wearable recording system to allow EEG recording over a 24 hour period (RFP-NINDS-73-15). This will assist in anti-epileptic drug evaluation in free-ranging patients in their natural environment, subjected to normal stresses. The Biomedical Engineering and Instrumentation Section will monitor and direct this prototype development project once successful contract negotiations are completed.

Ultrasound Techniques for Diagnosis of Cerebral Disorders:

The Biomedical Engineering and Instrumentation Section has planned and solicited proposals for a contract directed study of the feasibility of adapting ultrasonic techniques to provide detailed information about the status of brain tissue and other intracranial components which would allow a non-invasive diagnosis of specific pathologic states. The study will investigate the feasibility of proposed techniques and will determine the capabilities and limitations of sophisticated signal processing and instrumentation to differentiate pathologic states of interest in head trauma, stroke, TIA, multiple sclerosis, and cerebrovascular accidents on the basis of non-invasively measurable changes in the local mechanical properties of intracranial tissue. The Section will support and direct the project as soon as successful contract negotiations are completed.

EMI Scanner and Other X-Ray Tomography Studies:

The staff of the Biomedical Engineering and Instrumentation Section has, during the past year, participated closely in the Institute's investigation of the possible application and development potentials of x-ray tomography scanners such as the recently marketed EMI Scanner. Together with the Surgical Neurology Branch, NINDS, the Section has supported the design and

development , by BEIB/NIH, of a flexible high resolution phantom to evaluate the resolution capabilities of such scanners. This spring the Head of the Section will use this phantom to carry out such an evaluation of the EMI Scanner in London. Analysis of this data will allow for meaningful planning of clinical studies to be supported by other branches of NIH using these x-ray scanners for non-invasive assessment of intracranial disorders.

Laboratory Research and Feasibility Studies:

The Section carries out laboratory research aimed at evaluating instrumentation and techniques for their potential adaptation to specific research instrumentation needs of other programs of the Institute. Current activities include evaluation and development of EEG tape recording techniques, studies of optical scattering methods for antibody screening (NDS(CF)-73 BEI 2047) and development of rapid microspectrophotometry for study of photopigment kinetics. (Collaboration with the Laboratory of Neurophysiology, NINDS reported under projects NDS(1)-69 LNP/SP 1690 and NDS(1)-65 LNP/SP 1279).

Serial No. NDS(CF)-73 BEI 2047
1. Office of Associate Director
2. Section on Biomedical Engineering and Instrumentation
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
January 9, 1973 through June 30, 1973

Project Title: Biomedical Engineering and Instrumentation Studies

Previous Serial Number: None

Principal Investigator: George C. Murray, Ph.D.

Other Investigators: None

Cooperating Units: None

Man Years:

Total:	0.2
Professional:	0.2
Other:	0.0

Project Description:

Objectives: To provide the needed facilities and support for breadboarding and testing of instrumentation techniques and carry out feasibility studies and evaluation of possible application and adaptation of advanced technology to the measurement and instrumentation needs of Institute programs of neuroscience research and clinical neurology.

Methods Employed: Investigation of engineering data on potentially useful instruments and techniques and pilot studies to develop and evaluate needed design changes and test prototype approaches to problems related to the Section's instrumentation development programs.

Minimal laboratory and shop facilities are being developed for the Section staff to modify and test the capabilities of existing techniques and equipment for their potential development and application to specific instrumentation problems of the Section and other programs of the Institute. Wherever possible, cooperative work is carried out with the staff and facilities of the other labs and branches of the Institute and the other bioengineering facilities of the NIH.

Major Findings: By the end of the fiscal year instrumentation for light scattering measurements of antibody specimens will be completed and asymetry patterns should be determined and analyzed to determine design parameters for a simplified screening process.

Portable EEG tape recording equipment has been acquired and will be

evaluated against standard recording equipment. Designs for compression circuitry are being developed and will be incorporated by the end of the year.

Proposed Course of the Project: Continued investigation of antibody precipitate screening techniques to develop feasible approaches for instrumentation development.

Develop and pilot test methods for compression recording techniques to extend the useful dynamic range of magnetic tape recording of EEG.

Initiate and carry out a pilot study of other instrumentation, as need arises, in the development of new biomedical engineering programs to support the clinical research of the other branches of the Institute.

Honors and Awards: None

Publications: None

ANNUAL REPORT

July 1, 1972 through June 30, 1973

Applied Neurologic Research Branch
Collaborative and Field Research
National Institute of Neurological Diseases and Stroke
National Institutes of Health

The Institute's Collaborative and Field Research programs in Epilepsy, Head Injury, and Stroke were conducted by the Applied Neurologic Research Branch. Studies collaborating in-house research, directed (contract) research, and information programs were aided by support from the NIH Epilepsy Advisory Committee and its Subcommittees, and the NINDS Ad Hoc Committee on Cerebral Death.

Section on Epilepsy

Two laboratories within the Section participate in collaborative research activities with investigators not based at NIH. One of the laboratories provides closed circuit video monitoring of patient activity and simultaneous telemetry of electroencephalograms via FM tape recording through collaboration with the Departments of Neurology of the Medical College of Georgia and University of Virginia. Cooperation with investigators who employ computer-aided evaluation of electroencephalograms provided the capability of preparing compressed EEGs. This research tool will be useful in evaluating the effectiveness of therapy. Numerous technical problems are encountered in long term (12-hour) telemetry because of difficulty in maintaining integrity between the scalp and the electrodes. Consultation with NASA officials has offered some possibility of improved technology. The other laboratory is concerned with the pharmacology of anticonvulsant drugs and collaborates with the Medical College of Georgia, the University of Virginia, and the University of Kansas Medical Center. Other research is being conducted on the metabolism of investigational anticonvulsant drugs in animals. Some 10 technicians from laboratories throughout the country have visited the pharmacology laboratory to learn methodology and apply it in their labs.

The Subcommittee on Anticonvulsant Drugs of the NIH Epilepsy Advisory Committee (formerly the NINDS Ad Hoc Committee on Anticonvulsant Drugs) met in Bethesda in September 1972 and February 1973. The Subcommittee reviewed progress of NINDS sponsored research contracts and proposals for continued and additional studies. Modest programs were recommended in anticonvulsant drug synthesis and evaluation of the chronic toxicity of antiepileptic therapy.

Liaison activities with the pharmaceutical industry have continued. The Institute is currently sponsoring clinical evaluation of drugs from three companies, and several other firms have provided pharmacologic and clinical data on investigational anticonvulsant drugs. A double-

blind evaluation of clonazepam and ethosuximide in absence epilepsy are being investigated at the Medical College of Georgia and the University of Virginia. At New Castle State Hospital, chronic administration of combination treatment with carbamazepine, phenobarbital, and/or diphenylhydantoin in generalized seizures is under investigation. At the Lafayette Clinic, Detroit, the neurophysiological and neuropsychological parameters of carbamazepine treatment in partial seizures is under study. A double-blind study of sulthiame in outpatients with partial seizures was concluded at the University of Washington (Seattle). A study in the primate with a chronic lesion resulting in spontaneous seizures is employed to evaluate the relationship of blood levels of anticonvulsant drugs to seizure frequency and changes in behavior.

These collaborative studies entail cooperation between NINDS, University investigators, and liaison officials of pharmaceutical companies. Other pharmaceutical firms have sought advice from the Institute and its Subcommittee on Anticonvulsant Drugs before proceeding with development of their anticonvulsant compounds. Animal and clinical data has been reviewed and suggestions made to three companies about the best course for their research activities. Although the Institute is not sponsoring studies with compounds from these firms, it is possible in the future.

A review of the literature on the pharmacologic prophylaxis of posttraumatic epilepsy indicated that there is experimental and some clinical evidence that posttraumatic epilepsy may be prevented by prophylactic treatment with marketed anticonvulsant drugs. Unfortunately there has never been a large well-controlled clinical study to establish optimal preventive therapy. (Rapport RL, Penry JK: Pharmacologic prophylaxis of posttraumatic epilepsy--A review. Epilepsia 13:295-304, 1972.) Next, a survey of 1,064 board certified neurosurgeons was made to determine their current use of pharmacologic prophylaxis for posttraumatic epilepsy. Sixty percent of the respondents use anticonvulsants preventively, but under a variety of conditions. (Rapport RL, Penry JK: A survey of attitudes toward the pharmacologic prophylaxis of posttraumatic epilepsy. J Neurosurg 38: 159-166, 1973.) Thus, it was believed appropriate to undertake the pilot study to determine the effectiveness of diphenylhydantoin and phenobarbital in preventing seizures following injury to the head. The study is being conducted at the University of Kansas Medical Center as a guide to further studies. The use of nurse clinicians to assure followup is an important feature of this study. Also, blood levels of the anticonvulsants are employed to determine patient compliance and provide data for relation of drug level to effect.

Carbamazepine, an investigational anticonvulsant, is being evaluated in three separate clinical studies. This drug was chosen for intense investigation because of its wide use in Europe; also several hundred literature reports have appeared indicating effectiveness. The drug is marketed in the United States for the treatment of trigeminal neurolgia--thus it is available to those physicians who wish to employ it in epilepsy. However, carbamazepine has not yet been approved by the FDA for this indication. NINDS studies will aid in the evaluation of its

effectiveness and proper availability to physicians. At New Castle State Hospital, New Castle, Indiana, 30 patients are being treated for 1 year with carbamazepine, phenobarbital and/or diphenylhydantoin. This study is designed to determine long-term effects of carbamazepine combination treatment in a population of brain damaged patients with generalized seizures. At the Lafayette Clinic, Detroit, Michigan, a short-term study will be completed this summer to determine the effects of carbamazepine or placebo added to the treatment for patients with partial seizures with complex symptomatology. Preliminary results show that the drug is generally well tolerated and exerts an effect when taken along with the patients' other anticonvulsant drugs. There has been no obvious psychotropic effect noted and some mild behavioral toxicity has been observed. The investigators plan to continue the study on outpatients over a longer period of time. Patients with partial seizures with complex symptomatology have also been evaluated at the University of Washington (Seattle) where carbamazepine was employed as the sole anticonvulsant. The preliminary results are encouraging so that a long-term double blind comparison with diphenylhydantoin will be undertaken in this outpatient population. Careful monitoring of side effects, antiepileptic drug blood levels, and accurate reporting of seizures are important features of all of these studies.

The Congress has shown interest in providing funds to NINDS for the support of comprehensive epilepsy centers (CEC) where patient care, research, and rehabilitation are provided by referral of the patient with seizures. Although no money has been allocated for this purpose this year, a Subcommittee on Comprehensive Epilepsy Centers has developed the ideal attributes centers should possess. These principles will aid the Institute in developing feasibility studies and eventual funding for CEC.

The Section has continued to support the exchange of scientific information through several means. Excerpta Medica was provided support for the publication of Epilepsy Abstracts and its distribution to some 1,000 paid subscribers. In return, NINDS received the data base from Epilepsy Abstracts which may be searched through computers. The free text and retrieval system, Epilepsy Abstracts Retrieval System (EARS) was demonstrated and its value found high. Unfortunately, however, it was necessary to stop the feasibility study April 1, 1973. An epilepsy bibliography, 1900-1950, will be published mid-summer. It contains 12,000 citations and provides a compliment to the existing epilepsy abstracts which date from 1947.

NINDS Bibliography No. 2, Blood Level Determinations of Antiepileptic Drugs - Clinical Value and Methods, was published in fall 1972. Edited by J. K. Penry, L. D. Smith, and B. G. White, it provides authors and keyword indexes to 511 literature references on this subject. A motion picture, "The Absence Seizure," was produced in the Section's laboratory as a teaching tool for clinicians and others concerned with this disease of childhood. It has been viewed by numerous visitors to NINDS, at national meetings, and loaned to schools of medicine for instructional purposes.

Two books were published during the year by Raven Press. The first, Antiepileptic Drugs, edited by D. M. Woodbury, J. K. Penry, and R. P. Schmidt is a result of the 1971 symposium on this subject. Editorial assistance was provided by the Section. Experimental Models of Epilepsy--A Manual for the Laboratory Worker, edited by D. P. Purpura, J. K. Penry, D. Tower, D. M. Woodbury, and R. Walter was published in November 1972. Based upon a workshop held in 1971 and sponsored by the Subcommittee on Basic Research, NIH Epilepsy Advisory Committee, this book summarizes all present animal experimental models of epilepsy showing methods of preparation, and advantages and disadvantages of each technique. The scientific community has enthusiastically received these books. At its last meeting, the Subcommittee recommended that a workshop be held to discuss the neurosurgical treatment of epilepsy. This subject is considered very important for although there are limited indications for this form of therapy, it is effective under certain circumstances. Based on a conference sponsored by the Subcommittee on Epidemiology held in 1971, NINDS Monograph No. 14, Epidemiology of the Epilepsies, will be published this year. The monograph will show the state of the art and areas leading further study in the epidemiology of epilepsy.

In November 1972, with the Epilepsy Foundation of America and the American Epilepsy Society, NINDS sponsored a one-day workshop on laboratories for determination of antiepileptic drugs in serum. The workshop brought together persons who are establishing laboratories for measuring drug blood levels to define service requirements, applications, and limitations of various methods, the need for standards and research opportunities. The workshop achieved its purposes and was a helpful exchange of knowledge of these new, important techniques.

Section on Head Injury and Stroke

In June 1972, a brain death bibliography (BRAIN DEATH--A Bibliography with Key-Word and Author Indexes, NINDS Bibliography No. 1) was published by DHEW and received wide distribution through the biomedical research community.

The NINDS Collaborative Study of Cerebral Death continued gathering data throughout the year. By October, 500 patients had been entered and preliminary tabulations were complete on the first 401 cases. Delays in completion of checksheets at collaborating centers and submission to the coordinating centers produced a time lag in cross tabulation and data reduction. It was also quite clear that neuropathological studies were time consuming and expensive, yet had little correlation with either the clinical or electroencephalographic findings.

At the meeting of investigators and consultants of the NINDS Collaborative Study of Cerebral Death on October 19-20, 1972, it was decided to discontinue neuropathologic studies other than routine autopsies, and a slightly modified, simplified protocol was proposed for the remainder of the fiscal year. In addition, an editorial board was established in order to: (a) review manuscripts proposed for publication before the final report of the overall study is complete, and (b) write the final report.

By January 1973, the data indicated that of the patients admitted to the study with unresponsive coma and apnea, all but 27 died. Of 401 cases, 227 had electrocortical silence by EEG. The only survivors from that group were two cases of coma due to exogenous drug intoxication. The preliminary conclusion was, therefore, the triad of unresponsive coma, apnea, and electrocortical silence accurately prognosticated imminent death in 98 percent of cases. The 2 percent predictive error was caused by drug intoxications in which all elements of the triad are potentially reversible.

Furthermore, from a single blind analysis of admission blood samples done at the Pharmacology Laboratory of the Applied Neurologic Research Branch, more patients had measurable levels of intoxicating drugs than were suspected at the treatment centers. Therefore, it seemed important to develop another criterion for brain death which is independent of or insensitive to intoxicating agents.

Of the ancillary projects accomplished in the pilot study, the only additional test that still held promise by January 1973 was the bedside blood-flow assessment technic studied at New York University. The international literature on brain death suggested the technical feasibility of modified echoencephalography, evoked potentials, and compressed spectral analysis as alternate methods of increasing accuracy and/or objectivity in the prognosis of cerebral death.

The NINDS Ad Hoc Advisory Committee on Cerebral Death met on March 1-2, 1973, and reviewed the pilot phase of the study and the proposal for further collaborative studies. The Committee recommended that further investigation be concentrated upon the validation and field testing of the noninvasive bedside blood flow measurement and that all other aspects of the study be dropped. The Committee was very favorably impressed by the quantity and quality of data collected in the pilot phase and felt that the findings and conclusions should be communicated to the medical community as soon as possible. The Committee did not feel that the statistical conclusions would be significantly altered by adding to the sample size.

Plans for the next fiscal year include the continuation of data analysis and completion of the report based upon the total of 600 patients enrolled to date and for the further testing for accuracy, reliability, and validity of the technetium "bolus" technic of blood flow analysis. Completion of all phases of the Collaborative Study of Cerebral Death is expected in the next year.

In keeping with the suggestions of the Task Force on Head and Spinal Cord Injury, a directed research program on basic pathophysiology and applied rehabilitation of head injury was launched during fiscal 1972. A prospective study of psychological and vestibulo-auditory recovery after head injury was begun under contract with the University of Washington. Case finding methods and procedures were worked out during the first few months of the study. Through use of complete and comprehensive battery of vestibular and auditory function tests, the study quantitatively evaluates hearing and balance losses upon the recovery and rehabilitation of head-injured patients. This study is operating on schedule, and its goals and objectives will be accomplished within the negotiated contract period.

A basis research project began in the Departments of Engineering and Pathology at the University of Washington, testing the observed pathological changes in

cortical blood vessels and tissue after graded static loads are applied to exposed brain in rhesus monkeys. These experimental data are collated, compared and contrasted with predicted data from a computerized mathematical model of the stress fields created by blunt indentation of cortex.

The function of the Section was broadened to include directed program in certain areas of stroke research where more information is needed. The Task Force on Stroke pointed out that there were many unanswered questions regarding the character, frequency and prognostic significance of transient ischemic attacks. The data collection of the NINDS Cooperative Study of Hospital Frequency and Character of Transient Ischemic Attacks began in October 1972. Six collaborative centers have been contributing high quality pooled data with quarterly tabulations and continuous data analysis. Errors in case finding and coding were discovered and corrected as a result of continuing tabulation and data analysis. Although the study is descriptive and observational and is not intended to modify current treatment patterns, several trends have already indicated that this study will have far-reaching effects upon all future studies on the subject of TIA. For example, contrasting the enrollment rate in the latter half of the first year of the study to the earlier phase will indicate the true incidence rate of TIA (at hospitals) and the artifact produced early in the study by tapping the "pool" of patients reflecting prevalence plus incidence. Any incidence data derived from other prospective studies of treatment of TIA will have to take into account this "pool" effect. Of greater importance is the high rate (approximately 30 percent) at which the original diagnosis of TIA proved incorrect.

Work is in progress through a contract with the Ohio State Research Foundation to develop a new semiconductor gamma camera. This instrument when functional will offer a higher degree of spatial resolution than is possible with standard scintillation scanning equipment.

The Section on Head Injury and Stroke began planning for physical renovations for its in-house laboratory for the study of experimental models of cerebral ischemia and hemorrhage. During the renovation period, an interagency agreement was negotiated between the NINDS, C&FR program and the Armed Forces Radiobiology Research Institute which permits the experimental work of the laboratory to proceed during the period of laboratory renovation.

Regional and subregional blood flow monitoring methods are applied to specific models of cerebral ischemia and trauma in primates. Present work is concerned with the establishment of the time required for compensatory changes in circulation to occur without therapeutic intervention. Once this is established, therapeutic variables will be added, designed to salvage marginal zones of brain parenchyma which are at risk due to dysautoregulation. Pathophysiologic data from all experiments will be collated with qualitative and quantitative analyses of pathologic specimens.

The work of this laboratory has been planned to complement and interface with the contract-supported research programs of the seciton dealing with recovery of function after stroke and head injury.

CONTRACT NARRATIVE
Applied Neurologic Research Branch--Section on Epilepsy
July 1, 1972--June 30, 1973

NEW CASTLE STATE HOSPITAL (PH-43-NINDS-68-1310)

Title: Assess Carbamazepine as an Anticonvulsant Agent

Contractor's Project Director: Joseph T. Brock, M.D.

Current Annual Level: $73,143

Objectives: To study the relative anticonvulsant properties of carbamazepine, diphenylhydantoin, and phenobarbital administered to patients with seizures refractory to treatment, and to evaluate the possible side effects of the drugs and drug blood levels. The drugs are administered chronically in fixed combinations.

Course of Contract: Patient trials began September 10, 1972. Thirty-three patients began the one-year experimental design. The investigational drug carbamazepine was combined with two marketed drugs, phenobarbital and/or diphenylhydantoin. Clinical data was collected and sent to the Section on Epilepsy, NINDS, Bethesda, for review and preparation for computer-aided analysis.

Major Findings: Carbamazepine was found to have about the same anticonvulsant properties as diphenylhydantoin and phenobarbital when used singly in generalized seizures resistant to treatment. Combinations of the drugs were much more effective than single drugs. The present study will provide information about the relative effects of combined treatment.

Significance to NINDS Program and Biomedical Research: The pharmaceutical industry has demonstrated a little interest in developing new anticonvulsant agents. Aside from the economic factors involved, one of the industry's major problems is to obtain satisfactory clinical studies of anticonvulsant drugs. Through this contract and others, NINDS has supported clinical studies of antiepileptic drugs. Well controlled studies--as conducted at New Castle State Hospital for example--will be significant indicators of therapeutic merit of new antiepileptic drugs, and may encourage industry to develop promising agents for clinical trial. It is anticipated NINDS sponsored studies will enable carbamazepine to reach the market more readily, and thus be available to physicians who treat patients with seizures.

Proposed Course of Contract: Additional evaluations of antiepileptic drugs are planned.

Publications:

Cereghino JJ, Brock JT, Penry JK: Other hydantoins. Albutoin. In Woodbury DM, Penry JK, and Schmidt RP (Eds): Antiepileptic Drugs. New York: Raven Press, 1972, pp 283-291.

Cereghino JJ, VanMeter JC, Brock JT, Penry JK, Smith LD, White BG: Preliminary observations of serum carbamazepine concentration in epileptic patients. Neurology 23:357-366, 1973.

CONTRACT NARRATIVE
Applied Neurologic Research Branch--Section on Epilepsy
July 1, 1972--June 30, 1973

MEDICAL COLLEGE OF GEORGIA (NIH-NINDS-69-2169)

UNIVERSITY OF VIRGINIA SCHOOL OF MEDICINE (NIH-NINDS-69-2196)

Title: Study of Anticonvulsant Properties of Clonazepam and Ethosuximide

Contractor's Project Directors: Paul R. Dyken, M.D., (MCG)
Fritz E. Dreifuss, M.D., (UV)

Current Annual Level: $46,890 (Medical College of Georgia)
$65,907 (University of Virginia School of Medicine)

Objectives: To study and compare the effect of ethosuximide and clonazepam on the frequency and intensity of absence (petit mal) epileptic seizures in patients previously untreated for this disease and in patients who have failed to be controlled by ethosuximide; to evaluate the effect of ethosuximide and of clonazepam therapy on physiologic, psychometric, and other functions.

Course of Contract: One hundred patients will be studied, 50 at each center. Data is collected for each patient during the initial hospitalization period, during an outpatient and during the final hospitalization period, and sent to the Section on Epilepsy, NINDS, Bethesda, for review and entry for computer-aided analysis. One of the principal investigators changed locations during the year, and the study was transferred to his new location, the Medical College of Georgia.

Major Findings: The procedures and protocol established during earlier studies were proven valuable in conducting an evaluation of an antiepileptic agent. EEG telemetry has been developed as a major means of evaluating absence seizure frequency. Preliminary findings show that the two drugs evaluated have about the same effectiveness in absence epilepsy.

Proposed Course: The current study comparing ethosuximide with clonazepam will continue until the 100 patients described in the contract have been studied.

CONTRACT NARRATIVE
Applied Neurologic Research Branch--Section on Epilepsy
July 1, 1972--June 30, 1973

UNIVERSITY OF WASHINGTON (NIH-NINDS-70-2281)

Title: Study of Sulthiame in the Treatment of Partial Epilepsy

Contractor's Project Director: John R. Green, M.D.

Current Annual Level: $144,025

Objectives: To study the relative anticonvulsant properties in partial epilepsy of sulthiame and diphenylhydantoin; to measure these drugs in patients' blood, and to assess patients' psychological competence and social function. Also, to determine the effectiveness of carbamazepine in outpatients with partial seizures with complex symptomatology.

Course of Contract: The study was begun in October 1970; sixty-nine patients have been evaluated over a 14-month period. Twelve patients were studied in a preliminary evaluation of carbamazepine, as the sulthiame project was completed.

Major Findings: The contractor will review and analyze the data at the conclusion of the double-blind study. However, preliminary information indicates that sulthiame is less effective than diphenylhydantoin in the control of partial seizures.

Proposed Course: The contract will be continued with additional funds to compare carbamazepine and diphenylhydantoin in this patient population.

Publications:

Green JR, Kupferberg HJ: Sulfonamides and derivatives. Sulthiame. In Woodbury DM, Penry JK, and Schmidt RP (Eds): Antiepileptic Drugs. New York: Raven Press, 1972, pp 477-485.

Friel P, Green JR: Quantitative assay for carbamazepine serum levels by gas-liquid chromatography. Clin Chim Acta 43:69-72, 1972.

Martin SG, Green JR, Kehl TH, Amick-Corkill JA: Utilization of a microprogrammable computer for statistical analyses of data from a large epileptic patient population. I. Techniques and preliminary results. Epilepsia 13: 535-551, 1972.

CONTRACT NARRATIVE
Applied Neurologic Research Branch--Section on Epilepsy
July 1, 1972--June 30, 1973

UNIVERSITY OF WASHINGTON (NIH-71-2282)

Title: Study of Experimental Antiepileptic Drugs in Animals

Contractor's Project Director: Joan S. Lockard, Ph.D.

Current Annual Level: $60,682

Objectives: To compare the antiepileptic efficacy of carbamazepine, phenobarbital, and diphenylhydantoin in spontaneous motor seizures of primates. Seizure frequency and behavioral toxicity are compared with drug dosage and drug blood levels in the primate with spontaneous seizures.

Course of Contract: A pilot study in the primate revealed erratic absorption of carbamazepine and a very short half life. Primidone was substituted for carbamazepine so as not to detain the research.

Major Findings: The chronic lesion primate was found a suitable model for antiepileptic drug evaluation. The data obtained has not been analyzed, but is expected to show a relationship between drug blood level, EEG findings, and drug efficacy.

Proposed Course: The contract will be continued to further demonstrate and refine the usefulness of this animal model of epilepsy. Prophylaxis of posttraumatic epilepsy, motor seizure telemetry and social behavior, deep-sleep EEG telemetry and spontaneous seizures in the monkey model, and the efficacy of clonazepam and ethosuximide will be studied in the monkey model. The pharmacokinetics of clonazepam and ethosuximide will be determined in the primate before experimentation is begun.

CONTRACT NARRATIVE
Applied Neurologic Research Branch--Section on Epilepsy
July 1, 1972--June 30, 1973

LAFAYETTE CLINIC (NIH-NINDS-72-2310)

Title: Evaluation of Carbamazepine in Behavior and Physiologic Correlates of Temporal Lobe Epilepsy

Contractor's Project Director: Ernst A. Rodin, M.D.

Current Annual Level: $99,600

Objectives: To determine the effect of carbamazepine and placebo in 36 patients also treated with diphenylhydantoin and phenobarbital as measured by EEG telemetry and psychological measures.

Major Findings: Preliminary information shows carbamazepine exerts an effect when combined with diphenylhydantoin and phenobarbital; the previously reported "psychotropic effect" of carbamazepine has not been demonstrated.

Proposed Course: Following completion of this study of hospitalized patients, it is proposed to continue development of this study in outpatients.

CONTRACT NARRATIVE
Applied Neurologic Research Branch--Section on Epilepsy
July 1, 1972--June 30, 1973

UNIVERSITY OF KANSAS MEDICAL CENTER (NIH-NINDS-72-2313)

Title: Investigation of Posttraumatic Epilepsy Prophylaxis

Contractor's Project Director: Charles Brackett, M.D.

Current Annual Level: $92,050

Objectives: A pilot study to determine the effectiveness of prophylactic treatment with diphenylhydantoin and phenobarbital in persons who suffer head injury, and are thus liable to posttraumatic epilepsy.

Course of Contract: Although the admission rate is less than anticipated, there has been excellent patient rapport and compliance in the beginning months of the study.

Proposed Course of Contract: A sufficient number of patients will be studied to answer the hypotheses proposed and determine the necessity and feasibility for additional studies.

PARKE, DAVIS & COMPANY (NIH-NINDS-72-2315)

Title: Metabolic Disposition of Diphenylhydantoin

Contractor's Project Director: Anthony J. Glazko, Ph.D.

Current Annual Level: $93,754

Objectives: To determine the complete metabolic disposition in man of the antiepileptic drug diphenylhydantoin. Both oral and intravenous routes of administration will be followed, utilizing radioactive drug.

Course of Contract: Considerable time was required to obtain NIH human experimentation clearance for the project, as "normal" volunteers are studied. The radioactive drug was prepared, and the study began in March, 1973.

Proposed Course of Contract: The study will be concluded in the summer of 1973 and the contract extended without funds to permit data analysis.

CONTRACT NARRATIVE
Applied Neurologic Research Branch--Section on Epilepsy
July 1, 1972--June 30, 1973

EXCERPTA MEDICA FOUNDATION (NIH-NINDS-73-2303)

Title: Publication of Epilepsy Abstracts, Volume 6

Current Annual Level: $41,143

Objectives: To scan serial publications and periodicals from approximately 3000 of the world biomedical journals, select appropriate articles to be included in Epilepsy Abstracts in accordance with the guidance of the Project Officer and his editorial advisors; prepare abstracts with appropriate translations into English from foreign languages, classify, index, and store the abstracts in a computer retrievable form; and produce a 9-track computer tape in the NIH-PRS format, to be delivered when Volume 6 is completed. The Excerpta Medica Foundation will produce camera-ready copy for each monthly issue of Epilepsy Abstracts, which includes an index of subjects and authors, and will print and distribute the journal monthly, including a cumulative index at the end of the volume. In order to pay for the production of the camera-ready copy, the printing, and distribution, the Excerpta Medica Foundation sells subscriptions to recover the cost of production of camera-ready copy, printing, and distribution.

Course of Contract: Subscriptions to Epilepsy Abstracts, each at an annual cost of $20.00, have been acquired by Excerpta Medica at a satisfactory rate. Interest in the publication continues at a high level.

Proposed Course of Contract: It is anticipated that issues will be distributed as scheduled, and that the computer tape will be delivered in accordance with the contract. This tape will be added to the Epilepsy Abstracts Retrieval System (EARS) data base.

CONTRACT NARRATIVE
Applied Neurologic Research Branch--Section on Head Injury and Stroke
July 1, 1972--June 30, 1973

UNIVERSITY OF NEW MEXICO (NIH-71-2316)--Coordinating Center
VIRGINIA COMMONWEALTH UNIVERSITY (NIH-71-2315)
OHIO STATE UNIVERSITY (NIH-71-2317)
UNIVERSITY OF CALIFORNIA, SAN DIEGO (NIH-71-2318)
NEW YORK UNIVERSITY (NIH-71-2319)
NORTHWESTERN UNIVERSITY (NIH-71-2320)
UNIVERSITY OF UTAH (NIH-71-2322)
COLLEGE OF PHYSICIANS & SURGEONS-COLUMBIA UNIVERSITY (NIH-72-2312)

Title: Collaborative Study of Cerebral Death

Project Coordinator: A. Earl Walker, M.D. (UNM)

Project Directors: Cary Suter, M.D. (VCU)
Norman Allen, M.D. (OSU)
Reginald G. Bickford, M.D. (UCSD)
Julius Korein, M.D. (NYU)
Benjamin Boshes, M.D. (NU)
Donald R. Bennett, M.D. (UU)
Eli Goldensohn, M.D. (CPS)

Current Annual Levels: $94,377 (NM)
$48,240 (VCU)
$51,400 (OSU)
$27,000 (UCSD)
$73,000 (NYU)
$23,950 (NU)
$34,889 (UU)
$88,885 (CPS)

Objectives: The overall objectives of the collaborative study of cerebral death are 1) to verify or modify the current clinical and electroencephalographic criteria for cerebral death in relation to patient age and cause of coma; 2) to determine the minimal time that clinical and EEG criteria must be operative to indicate cerebral death in relation to age and cause of coma; 3) to assess the neurologic deficit of patients who improve after having fulfilled all criteria for some length of time; and, finally, 4) to characterize abnormal EEG patterns seen prior to electrocerebral silence.

Major Findings: By November 30, 1972, 500 cases had been entered into the study. Data analysis which had been begun at that time is still ongoing to cross correlate the many variables identified in the study. An interim simplified protocol has been adopted which is addressed to the principal variables identified in the pilot phase.

The general results have shown that, except for cases of drug intoxication, a one-hour flat EEG accurately predicts a cardiac death. There have been two cases of drug intoxication (2%) in which the EEG's have been flat and subsequent recovery achieved. In view of this fact, any future studies will have to provide an alternative means of identifying the drug-intoxicated cases.

The study has shown that either by history or by lab test, significant errors may occur in identification of the drug-intoxicated cases. Furthermore, the dose response relationship between drug level, ECS and recovery from coma has varied. Since adequate qualitative and quantitative drug screening procedures are not yet in general use, some measure of cerebral blood flow seems the most feasible method for identification of patients with reversible electrocortical silence.

Neuropathological studies have shown less than perfect correlation between pathologically dead brains and electrocortical silence. In view of this fact, future studies will be limited to routine autopsy information and the detailed neuropathology protocol will not be continued.

Based upon operational experience, the ophthalmologic examinations, the compressed spectral array analysis, the serial CSF determinations, and the dopamine beta hydroxylase studies will not be continued in the future work of this study.

The only ancillary study which holds promise is that of the rapid intravenous injection of technetium^{99} with single probe scintillation counting over the skull. This "bolus" technic is possibly a representation of cerebral blood flow. More tests are needed in order to validate this technic and to test its reliability in patients with ECS as a predictor of death. The hypothesis is that a "bolus" effect will be present in patients who have ECS due to drug intoxication and will indicate at least marginal cerebral blood flow.

Significance to NINDS Program and Biomedical Research: Considerable controversy still exists over the legal, moral, and scientific definitions of death. The criteria used for the diagnosis of brain death vary in the world literature. The need for reliable uniform criteria still exists in order to spare families the emotional stress and financial burden of unnecessary delays in pronouncement of death, and, at the same time, to protect against undue haste in cessation of efforts of resuscitation. European criteria include assessment of blood flow by aortocranial angiography in primarily brain-damaged patients while the Harvard Criteria which do not require blood flow assessment, remain the most widely accepted norms in this country. The results of this study may modify the Harvard Criteria which require the exclusion of patients with drug intoxication and hypothermia.

Proposed Course of Contracts: Some future contracts may be negotiated dependent upon prospective contractors' ability to fulfill recommendations of the handling of the high priority work identified in the pilot phase, i.e., to develop a valid, noninvasive method for assessment of blood flow to the brain in unresponsive comatose patients with electrocortical silence.

CONTRACT NARRATIVE
Applied Neurologic Research Branch--Section on Head Injury and Stroke
July 1, 1972--June 30, 1973

OHIO STATE UNIVERSITY RESEARCH FOUNDATION (NIH-72-2323)

Title: To Develop a New Semiconductor Gamma Camera System for Nuclear Medicine

Contractor's Project Director: Robert F. Redmond, Ph.D.

Current Level: $165,538 (18 months)

Objectives: The objectives of this contract are to design, build, and evaluate a prototype semiconductor gamma camera for dynamic cerebral vascular studies in humans based on a position sensitive germanium detector.

Major Findings: Two major areas of investigation were selected; the development of semiconductor technology which is suitable to the fabrication of germanium position sensing detectors and the development of appropriate signal processing techniques and electronic instrumentation which will result in camera resolution on a millimeter scale. To date, progress of the work has been satisfactory. Germanium detectors with resistive electrodes formed by ion implantation have been fabricated and their properties measured. Significant progress has been made in the evaluation and fabrication of electronic signal processing equipment and the optimization of camera system design. The prototype of the electronic signal processing system is under construction. Computer codes have been completed which are capable of theoretical calculations of the system's performance and will be compared with the measured characteristics of the camera under actual operating conditions.

Significance to NINDS Program and Biomedical Research: The gamma camera produced from the above contract should allow increased spatial resolution of lesions which are identifiable by radiopharmaceutical uptake. Such refinements in scintillation scanning will complement recent advances in x-ray technology, such as transverse axial tomography.

Proposed Course of Contract: The progress of developmental work has been on schedule and completion is expected by the termination date. Management responsibility for this contract will be transferred to the Section on Bioengineering.

CONTRACT NARRATIVE
Applied Neurologic Research Branch--Section on Head Injury and Stroke
July 1, 1972--June 30, 1973

INDIANA UNIVERSITY FOUNDATION (NIH-72-2324)

Title: A Study of the Hospital Frequency and Character of Transient Ischemic Attacks--Mark L. Dyken, M.D., Principal Investigator

Current Level: $196,781 (18 months)

Collaborating Centers: University of Maryland
University of Mississippi
Institute of Medical Sciences
University of Washington
Massachusetts General Hospital
Indiana University

Project Directors: Thomas R. Price, M.D. (UMd)
Armin F. Haerer, M.D. (UMiss)
Philip R. Calanchini, M.D. (IMS)
Phillip D. Swanson, M.D. (UW)
David C. Poskanzer, M.D. (MGH)
Mark L. Dyken, M.D. (Ind)

Objectives: The objectives of this study are to determine prospectively the relative frequency of each of the etiologies of transient ischemic attacks and to describe the current standard of diagnostic work-up, diagnostic accuracy, and therapy. The study is limited to patients presenting for hospitalization and hospital outpatient clinics.

Major Findings: In the first four months of data collection, 174 pateints had been entered into the TIA collaborative study. Up-to-date processing by the Indiana group allowed a rapid evaluation of our results so far. A "pool" effect has been demonstrated as an artifact in prospective study design. In the first few months of case finding, the admission rate to the study reflects a summation effect of prevalence plus true incidence rates in the patient population. Preliminary analysis suggests a higher than predicted rate at which initial diagnoses of TIA are proved incorrect after work-up.

Significance to NINDS Program and Biomedical Research: A data base for evaluating the character, frequency and short term prognostic significance of transient ischemic attacks will be provided through this project. The findings of this study when completed will influence the interpretation of data from past and ongoing studies of the epidemiology and therapy of TIA.

Proposed Course of Contract: The true significance of the pool effect and the short term outcome after well documented TIA's will require a limited extension of data gathering and a follow-up period.

CONTRACT NARRATIVE
Applied Neurologic Research Branch--Section on Head Injury and Stroke
July 1, 1972--June 30, 1973

UNIVERSITY OF WASHINGTON (NIH-72-2325)

Title: Fragility of Brain Tissue

Contractor's Project Directors: James D. Chalupnik, Ph.D.
E. C. Alvord, Jr., M.D.

Current Annual Level: $49,990

Objectives: The purpose of this project is to establish a fragility index for brain tissue in vivo, reflecting the minimum levels of stress and strain likely to cause histological damage. This is to be accomplished by comparing experimental data derived from animals to predicted data from a computerized mathematical model of static loading of cerebral cortex.

Major Findings: The computer program development which is to predict the strain states under various depths of indentation is continuing but is approximately two months behind schedule. To date there are no useful data reported from animal model studies.

Significance to NINDS Program and Biomedical Research: As originally designed, this study is to use data from the Head Injury Model Project, and the findings should complement that study. The instrument used to generate the stress fields in brain tissue can provide a predictable field but does not simulate the blunt trauma caused by accidental head injury in humans. To establish norms of microvascular fragility remains an important goal which this project could achieve.

Proposed Course of Contract: Delays in equipment design and manpower shortages have put the experimental animal studies far behind schedule. The project will be extended beyond the original termination date without additional funding contingent upon satisfactory evidence that the project can be completed in six months time.

CONTRACT NARRATIVE
Applied Neurologic Research Branch--Section on Head Injury and Stroke
July 1, 1972--June 30, 1973

UNIVERSITY OF WASHINGTON (NIH-72-2326)

Title: Human Responses to Head Injury

Contractor's Project Director: Ralph M. Reitan, Ph.D.

Current Level: $199,713 (24 months)

Objectives: This contract is designed to investigate the recovery of balance and cognitive functions following significant head injuries. Patients between ages 15-50 years of age who have suffered head injury with 1) unconsciousness for at least 10 minutes or 2) objective evidence of localized cerebral trauma will be examined for one year following the injury. Follow-up tests include time-locked studies of comprehensive neurophysiological testing, complete audiologic and vestibular testing, electroencephalography, and clinical neurological evaluations. The complete test battery will be repeated at three-month intervals during the year, following the patient's admission to the study.

Major Findings: A trend has been identified suggesting seasonal variation in the incidence of head injuries. Among the first 17 patients admitted to the study, absence of caloric responses in some who are given Dilantin and unusual eye-tracking findings during vestibular testing have been documented. The statistical significance of these findings remains to be established as the sample size increases. The prognostic significance will be determined by the testing during the follow-up period.

Significance to NINDS Program and Biomedical Research: The psychological and hearing abnormalities developing after head injury are difficult to manage in current practice. Hearing loss and receptive dysphasia may be present as confusing constellation of behavioral abnormalities early in the recovery period. There are often months to a one-year delay from the time of injury to adequate assessment of symptomatology. Dysequilibrium caused by vestibular damage impedes the recovery of independent ambulation. This research is expected to provide useful information in forecasting possible vestibular and psychological changes following head injury and contribute to the planning of effective rehabilitation programs.

Proposed Course of Contract: Early operational problems in case finding and vestibular test equipment failures have been corrected. The two-year study is planned to continue until completion with no additional contracts planned at this time.

Serial No. NDS (CF)-71 AN 1932
1. Collaborative and Field Research
2. Epilepsy Section
3. Bethesda, Maryland

PHS--NIH
Individual Project Report
July 1, 1972--June 30, 1973

Project Title: Long-term electroencephalographic telemetry of patients with absence (petit mal) seizures.

Principal Investigator: J. Kiffin Penry, M.D.

Other Investigators: Stephen W. Rose, M.D.
Fritz E. Dreifuss, M.D.
Paul R. Dyken, M.D.

Cooperating Units: Departments of Neurology, University of Virginia School of Medicine, Charlottesville, Virginia, and Medical College of Georgia, Augusta, Georgia.

Man Years:

Total:	0.75
Professional:	0.25
Other:	0.5

Facilities and Equipment:

The telemetry equipment and FM tape recorder were provided by the Section on Epilepsy. The clinical research units were provided by the University of Virginia Hospital, and the Medical College of Georgia Hospital.

Project Description:

Objectives: This study evaluates patterns of paroxysmal abnormal discharge in patients with absence (petit mal) seizures. Another primary objective is to determine if this method can be established as a primary mode of evaluating any absence drug.

Methods employed: Each patient has a 12-hour telemetered electroencephalographic recording on the clinical research unit. The telemetry equipment is placed on the patient's head by an EEG technician who also applies the electrodes. The patient is free to move about the ward and the signal from the transmitters is taken to a dipole antenna and then to four receivers where four channels of the EEG are fed to an FM tape recorder. Quality control is maintained by observation of the incoming data on the oscilloscope. The 12-hour recordings are taken back to the laboratory of the Epilepsy Section in Bethesda where they are played back at four times recording speed. Each 12-hour record is then read by a physician. Patterns of discharge and effects of anti-absence drugs are evident when the data is processed by a 360 IBM computer and the Calcomp plotter.

Major Findings: Patients with absence epilepsy have generally shown improvement following treatment with absence drugs. Many patients have shown complete remission of paroxysmal activity. It was felt that this criterion of seizure control is probably the most objective available, and it is the intent of the Section on Epilepsy to incorporate this method in evaluation of investigational anti-absence drugs.

On further evaluation, 16 patients with absence seizures have shown a decline in seizure duration with increasing age. While this is a confirmation of previously suspected phenomena, this is the first time that such information has been collected in other than an anecdotal way.

Significance to Biomedical Research and to the Program of the Institute: This study has applied telemetry techniques to improve the evaluation of clinical research in anti-absence pharmacology. The study has further suggested patterns of paroxysmal discharge in patients with absence seizures and points a way for further investigation of the mechanisms of absence epilepsy.

Honors and Awards: None

Publication:

Browne TR, Penry JK, Porter RJ, Dreifuss FE: Responsiveness before, during, and after spike-wave paroxysms (abstract). Neurology 23:403, 1973.

Serial No. NDS (CF)-71 AN 1933
1. Collaborative and Field Research
2. Epilepsy Section
3. Bethesda, Maryland

PHS--NIH
Individual Project Report
July 1, 1972--June 30, 1973

Project Title: Quantitation of clinical manifestations of spike-wave activity by a reaction time method.

Principal Investigator: J. Kiffin Penry, M.D.

Other Investigator: Fritz E. Dreifuss, M.D.

Cooperating Units: Department of Neurology, University of Virginia School of Medicine, Charlottesville, Virginia.

Man Years:

Total:	0.5
Professional:	0.25
Other:	0.25

Facilities and Equipment:

Electroencephalographic equipment and space for testing were provided by the University of Virginia Hospital at Charlottesville. The reaction time equipment including seizure detection device and digital timer were designed and built by the Section on Epilepsy. The video-recording apparatus was provided by the Section on Epilepsy.

Project Description:

Objectives: The purpose of this study is to determine whether reaction time in absence patients is or is not impaired in a gradual fashion from the point of spike-wave initiation as has been suggested by some authors but disputed by others. There is some evidence for a "trough-like" pattern decrease of consciousness. The onset of decrease clinical functions during spike-wave paroxysms is evaluated by the reaction time method.

Methods employed: A device is employed which gives instantaneous recognition by voltage criteria that a spike-wave burst has started. This burst is of much higher than normal background, and this factor alone is used to electronically trigger the reaction timer. On instantaneous recognition the reaction timer is triggered and a tone is delivered to the subject. The subject responds by turning off the high pitch tone with a telegraph key. Between paroxysms the patient is maintained in a state of alertness by a program of approximately 10 random stimuli per minute. All the data is collected by television, including a portion of the screen reserved for the reaction time from the digital clock. There is no age limit in selecting patients,

but they must all have spike-wave paroxysmal discharge. A second group of patients was studied with the apparatus altered slightly so that the auditory stimulus was delivered 0.5 seconds into the seizure in order to see if responsiveness becomes less as the seizure progresses.

Major Findings: The findings of the first group of patients suggest that some ability to respond early during the paroxysmal burst is maintained; this responsiveness is frequently not seen 1-2 seconds after onset. Analysis of responsiveness during short bursts suggests that patients may retain a normal reaction time during such paroxysms.

Significance to Biomedical Research and to the Program of the Institute: This study has applied video recording techniques and sophisticated electronic methods to improve the quality of clinical research. Specifically, this study is an analysis of the relation of the patient's behavior to his EEG during paroxysmal electroencephalographic events. An understanding of this relationship is important--not only as a guidepost for further research in the mechanism of epilepsy, but also in determining the day-to-day therapeutics of the epileptic patient.

Proposed Course: The study will be continued with additional patients in the coming fiscal year.

Honors and Awards: None

Serial No. NDS (CF)-73-AN-2049
1. Collaborative and Field Research
2. Epilepsy Section
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972--June 30, 1973

Project Title: A Radioimmune Assay for Diphenylhydantoin

Principal Investigator: Richard L. Rapport, II, M.D.

Other Investigators: Robert E. Tigellar, M.D., John K. Inman, Ph.D., Harvey J. Kupferberg, Ph.D.

Cooperating Units: Laboratory of Microbial Immunity and Laboratory of Immunology, NIAID.

Man Years:

Total:	0.25
Professional:	0.15
Other:	0.1

Facilities and Equipment:

Laboratories of the three cooperating units at the National Institutes of Health were employed.

Project Description:

Objectives: To devise an original method for the assay of antiepileptic drug diphenylhydantoin (DPH) in blood using radioimmunoassay methodology. This technique offers advantages over other methods of assay, especially that the amount of blood required may be obtained from a "finger stick," or other small quantities.

Methods employed: Anti-sera were prepared by immunizing rabbits with a DPH-chicken gamma globulin conjugate. The assay is based upon the competition for binding the anti-DPH antibodies between a standard of labeled DPH and unlabeled DPH.

Major findings: The assay can detect as little as 0.03 ug of DPH in 0.1 ml sample. The metabolite parahydroxyphenyl phenylhydantoin (HPPH) reacts as well as DPH, but other hydantoin antiepileptic drugs and phenobarbital do not. Parallel analysis by radioimmune assay and by gas-liquid chromatography produced excellent correlation between the two methods.

Significance: An alternate method of antiepileptic drug assay has been developed; it can be done quickly and reliably in laboratories capable of radioimmune assays.

Proposed Course: The study has been completed.

Publication:

Tigellar RE, Rapport RL II, Inman JK, Kupferberg HJ: A radioimmune assay for diphenylhydantoin. Clin Chim Acta 43:231-241, 1972.

Serial No. NDS (CF)-73-AN 2050
1. Collaborative and Field Research
2. Epilepsy Section
3. Bethesda, Maryland

PHS--NIH
Individual Project Report
July 1, 1972--June 30, 1973

Project Title: Metabolism of Dimethoxymethyl Phenobarbital (DMMP) in Mice

Principal Investigator: Harvey J. Kupferberg, Ph.D.

Other Investigators: Richard L. Rapport, II, M.D.
Wayne D. Yonekawa

Man Years:

Total:	0.5
Professional:	0.3
Other:	0.2

Project Description:

Objectives: To investigate the relationship between the anticonvulsant activity of DMMP--an investigational drug--and its metabolism to phenobarbital in mice. Other N alkylated substituted anticonvulsants are dealkylated in vivo and the metabolites are equally or more active than the parent drug. Thus, it is appropriate to investigate the new agent, DMMP, for similar activity.

Methods employed: DMMP and phenobarbital were administered intragastrically to fasting mice and the time of peak anticonvulsant effect determined. Brain, blood, and whole body phenobarbital levels were determined following drug administration, using gas-liquid chromatography.

Major Findings: Brain levels of phenobarbital 3 hours after administration of DMMP averaged 24.6 ug/ml; following an equivalent dose of phenobarbital levels averaged 25.4 ug/ml. Blood and whole body phenobarbital levels paralleled those in brain. The data shows that after a single dose of DMMP in mice, its anticonvulsant activity measured by electroshock protection is a result of its metabolism to phenobarbital. Monomethoxymethyl phenobarbital was isolated following large doses of DMMP. This intermediate was found to have activity against electrically induced seizures.

Significance to Biomedical Research and to the Program of the Institute: The metabolism of investigational anticonvulsant drugs is important to their further development. It is only after this information is available that further animal studies can be conducted and consideration given to clinical trial for efficacy if promise is shown.

Proposed Course: The study has been completed.

Publication:

Rapport RL, Kupferberg HJ: Metabolism of dimethoxymethyl phenobarbital (DMMP) in mice: Relationship between brain phenobarbital levels and anticonvulsant activity. J Med Chem, in press.

Serial No. NDS (CF)-73 AN 2051
1. Applied Neurologic Research Branch
2. Section on Head Injury and Stroke
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
April 1, 1973 through June 30, 1973

Project Title: Experimental models laboratory--head injury and stroke

Previous Serial Number: None

Principal Investigator: G. F. Molinari, M.D.

Other Investigators: John I. Moseley, M.D. (NINDS)
J. Fein, M.D. (AFRRI)

Man Years:

Total:	1.5
Professional:	1.0
Other:	0.5

Project Description:

Objective: To establish the maximum duration of cerebral tolerance of ischemic anoxia and to evaluate the association of ischemia and hemorrhage in primate models of head injury and stroke.

Methods Employed: Embolism and transorbital clipping of middle cerebral arteries are to be compared and contrasted for size, distribution and consistency of gross and histopathologic lesions. Having established the pathologic anatomy of these models, acute experiments will employ hydrogen washout blood flow method and EEG to study the early stages of evolution of ischemia and infarction. These parameters will be monitored simultaneously in both primary and secondary zones of disordered circulation.

Major Findings: Models have been developed for segmental cerebral artery occlusion, cerebral vasospasm, and closed head injury. Hemorrhagic or bland infarction can be selectively produced.

Significance to Biomedical Research and the Program of the Institute: These experiments are designed to provide basic pathophysiologic data related to secondary prevention as applied to stroke and head injury. While certain areas of direct traumatic necrosis or total cessation of blood are inevitable, functional sequellae may be minimized by prevention of detrimental secondary vascular events in fields of collateral circulation.

These experiments are expected to complement clinical studies of secondary prevention in patients supported by the Section's field research program.

Proposed Course: With an Interagency Agreement between National Institute of Neurological Diseases and Stroke, NIH, DHEW, and The Armed Forces Radiobiology Research Institute Defense Nuclear Agency (subject to renewal), the Section on Head Injury and Stroke will continue its laboratory investigations through this agreement until completion of the "in house" laboratory renovation, targeted for late 1973. At that time, stroke model investigations will be performed in the section laboratory, NINDS. Continuation of head injury model work and certain blood flow measurements will be continued at AFRRI subject to renewal of the agreement.

ANNUAL REPORT
JULY 1, 1972 THROUGH JUNE 30, 1973
EPIDEMIOLOGY BRANCH
COLLABORATIVE AND FIELD RESEARCH
NATIONAL INSTITUTE OF NEUROLOGICAL
DISEASES AND STROKE

Introduction

The major areas of activity of the Epidemiology Branch during the period of this report have been:

I. Epidemiologic studies of neurologic diseases

II. Laboratory studies related to the epidemiology and immunology of neurologic and perinatal diseases

III. The continued activities on Guam and the Trust Territories

IV. Genetic studies of neurologic diseases

The permanent professional personnel of the Epidemiology Branch has not changed during the year. Included are Jacob A. Brody, M.D., Branch Chief, Dwayne Reed, M.D., Deputy Branch Chief, Roswell Eldridge, M.D., Head, Section on Genetics in Epidemiology, and in the Laboratory, Lon White, M.D., and George Nemo, Ph.D. (Virology). Mr. Stephen Showalter's temporary appointment in the laboratory expired in February, and we are attempting to replace him since Dr. Nemo has no technical assistance at present. Dr. Milton Koch joined the Epidemiology Branch staff in July 1972.

In October of 1972, the Branch was moved, leaving the laboratories in Building 36, while the Epidemiology and Genetics groups were relocated on separate floors of the Federal Building. In February 1973, Dr. Reed was assigned to Guam for one year to complete the studies requested by the Guam Peer Review in February 1972. The Branch had a Peer Review in late March and another Peer Review is scheduled for mid-July. In addition, a Peer Review is scheduled for the Guam projects in September of 1973. We have contributed heavily in Institute and Division organizational activities and have prepared state of the art papers on amyotrophic lateral sclerosis and Parkinson's disease and have served on Task Teams for Multiple Sclerosis and Communicative Disorders.

I. Epidemiologic Studies of Neurologic Diseases

Multiple Sclerosis

We remain active in the field of multiple sclerosis, with in-house studies, a contract with the NAS,direct participation of the National Multiple Sclerosis Society, and the C&FR MS Task Team. In addition, we were called upon to discuss "Epidemiologic and Serologic Data on Multiple Sclerosis and Their Possible Significance" at the most recent International Multiple Sclerosis Symposium. (UCLA Forum and Medical Sciences #16, Multiple Sclerosis, Immunology, Virology and Ultrastructure).

Our in-house studies are directed toward determination of environmental factors which could explain the geographic distribution of MS and the observation that critical determinants in acquiring subsequent MS occur before age 20. We now know of 5 apparent foci of MS, one in Comanche County, Kansas, a second in Mossyrock, Washington, a third in Superior, Wisconsin, a fourth in Danbury, Connecticut, and a fifth in suburban Boston. We have partially worked up the first two and intend to continue with descriptive studies of the other areas, in hopes of finding common environmental factors. In Comanche County we know of 7 cases in a population of 3,000, and are in the process of writing to people who migrated from the area. In this manner, we have picked up at least 2 more patients which increases the potential significance of this cluster. In Mossyrock, Washington, we learned of 6 patients in 3 families, all born between 1916 and 1920, and an additional unrelated patient born in 1940. Of possible significance, is the fact that a smallpox epidemic occurred in this area in 1924, and in the same year there were 10 infant deaths listed as being caused by measles. In the region around Wichita, Kansas, we are in the process of studying a large series of patients and controls who were born in that area. We are concentrating on the sequence of infections and exposures which occurred before age 20, through questioning the mothers of patients and controls in a detailed manner. As would be expected, we are having difficulty finding enough patients and controls whose mothers are available for interview. We expect however, to have enough cases for analysis within the next year. We have designed protocols and are in contact with the National Multiple Sclerosis Society and the Department of Social and Preventive Medicine of the University of Dundee in Scotland, in order to conduct an ecological study of the Shetland and Orkney Islands, where we confirmed that MS is occurring at a rate 3 times higher than that reported anywhere else in the world. Our contract with NAS is for analysis of patterns among 6,000 veterans with MS. (See details under Contracts). Laboratory investigations of MS are described in Section II of this summary.

Amyotrophic Lateral Sclerosis

In addition to our extensive studies on Guam, we have a series of in-house studies, a contract with the NAS, and participation with the Muscular Dystrophy Associations of America, which funds research in motor neuron diseases. In addition, we have prepared an extensive review of

the state of the art, and recommendations for areas of possible studies of ALS.

Following up on our observation of ALS patients on Guam and in the United States having depressed CNS dopamine metabolism, we have collaborated with Dr. Andre Barbeau, Director of Department of Neurobiology, Clinical Research Institute of Montreal, in the analysis of fresh brain material of ALS patients. Preliminary results indicate that there is no cerebral dopamine metabolism deficiency at the cellular level in ALS.

Analysis of deaths among 12,000 statesiders who worked for more than one year on Guam between 1945 and 1955 has been updated, and we are awaiting several computer analyses. It does not appear however, that these individuals were at increased risk of developing ALS. In view of several reports that pancreatic insufficiency may be associated with ALS and other neurologic diseases, we have once again updated our series of peptic ulcer patients in a large VA study in which a portion of ulcer patients were treated with surgery which interfered with pancreatic function. We are now analyzing information on some 2,000 patients. Material has been coded and we are awaiting the computer runs. Our impression is that there was not an excess of ALS but there may be an excess of severe neurologic disease in general in the operated population.

Our contract with NAS involves studying motor neuron disease among veterans. The main purposes are 1) to determine risk factors based on 700 patients and matched controls, 2) to determine the natural history of the various forms of motor neuron disease in approximately 300 well documented patients, 3) an extensive questionnaire of 200 living ALS patients matched with 200 living patients with brain tumors. The study is in its second year and still in the data collection phase.

Stroke

We have small in-house studies of stroke and several large contracts. Data from several of our previous contracts indicate that hypertension and cardiovascular disease occur in a distinctive geographic pattern in the United States, being highest in the southeastern states and lowest in the mountain states, with the remaining states intermediate. In collaboration with the Epidemiology Branch, NIHL, we are studying patterns of stroke among resident and migrant populations in California. This state has a very large migrant population and approximately 16,000 stroke deaths per year. We are in the process of negotiating with the California State Health Department for data for the two 3-year periods around the 1960 and 1970 census. Analysis should elucidate whether migration away from a high risk area or a low risk area to an intermediate risk area affects the risk of developing a stroke and should be useful in determining the age at which environmental factors appear to exert their influence.

Through a contract with the School of Public Health at the University of Texas and a subcontract to the University of Panama, we are studying stroke risk factors in selected black populations to determine the relative role of environment and heredity in the observed higher rates of stroke among American blacks. Through a contract with the Kaiser Permanente in San Francisco, we are studying a population which developed a stroke at some time after a multiphasic screening exam. This population will be compared with the general population and also with subpopulations having similar known risk factors for stroke, but who did not develop a stroke in the ensuing interval. By controlling for risk factors new and more subtle and potentially important risk factors may emerge.

Through a previous contract, we documented that stroke and hypertension occur among Chamorros on Guam and in California at approximately the same rate as they do in the general United States population. We have neuropathologic observations however, that atheromata occur at a much lower rate in the vessels of the brains in the adult population of Chamorros on Guam. This and the fact that we have an accessible resident population with large kindreds could lead to potentially important studies of families with and without stroke. At present we are collecting some statistical information on stroke on Guam, but do not have the staff or funds to enter into a definitive study.

Parkinson's Disease

The great improvement in symptoms of Parkinson's disease patients with the drug L-Dopa has rapidly led to its wide acceptance. The long term effects of L-Dopa on the natural history of Parkinson's disease and possible harmful effects of taking this medication over many years has not been determined. While a case-control study would be the optimum way of determining the long-term effects of L-Dopa it would not be ethical at the present time to withhold the drug from a large number of cases. We have therefore been trying to identify populations in which there was sufficient information on a pre-L-Dopa cohort to determine a life table which could be compared with a cohort on L-Dopa. We have worked with the National Parkinson Foundation, Inc., in Miami and with the New York Hospital but in each case either follow-up has proven impossible or self-selection of patients in the pre-L-Dopa cohort made comparison with the post-L-Dopa cohort invalid. During the past year we have been attempting to determine if a valid pre- and post-L-Dopa cohort could be identified in Saskatchewan. The province of Saskatchewan began a socialized medicine program in 1962, with the unique feature of having a central recording system. L-Dopa was not used until 1970. The overall population is one million with a relatively high proportion in older age groups. Thus, it might be possible to utilize this population for a suitable study. At present, we are working with the neurologist at the University of Saskatchewan School of Medicine. Various problems of medical confidentiality and access to data have been eliminated. Hopefully a pilot study will be initiated in the near future with funding

from Canadian Government sources. We are awaiting the outcome of a research grant proposal. If this is unsuccessful I believe we should explore other mechanisms whereby we can ascertain the feasibility of this study in Saskatchewan.

Infections or Possible Infections

Subacute Sclerosing Panencephalitis:

In our previous study we demonstrated that SSPE frequently developed in children who get clinical measles under age 1 or have inapparent measles infections or measles infections associated with concomitant chickenpox. These unusual situations apparently are involved with measles becoming a chronic infection. Several other groups have now developed animal models along these lines for the establishment of chronic measles virus infection. Our data further suggested that a second triggering phenomenon is probably involved since SSPE occurs primarily in non-urban males. One lead in our small series was the exposure to clinically diagnosed distemper, a virus related to measles in the families of the patients. In collaboration with the Division of Biologic Standards, we are attempting to develop a satisfactory serologic test for distemper to use in our patient and control material, and possibly in ecologic studies in areas where cases occur.

Creutzfeldt-Jakob Disease:

Our analysis of a case-control study of 66 patients is now complete. We found that patients with the classical or slowly progressive form of the disease associated with amyotrophy are on an average 10 years younger than other CJD patients. This type of disease has not yet transmitted to chimpanzees. Severe upper respiratory illness merged with the onset of CJD in 3 patients with the ataxic form of the disease. No specific factors, exposures, or prior history of disease distinguished the patients from the controls. It was of interest however, that one-third of the patients and one-third of the controls ate animal brains. The patients tended to have eaten animal brains more frequently and more recently than controls. Also 10 of the 13 patients and only 3 of the controls stated specifically that they ate hogs' brains. While there was no overall difference between patients and controls, this may be a lead since the other slow virus transmissible spongiform encephalopathies are transmitted in nature through exposure to brain material. We are now extending our study by using a modified questionnaire in additional patients and controls to get more information on consumption of animal brains and respiratory illness. We are also trying to secure information on animal brain consumption in the United States and Great Britain.

Juvenile Polymyositis:

We are conducting a case-control study of juvenile polymyositis using an extensive questionnaire. We have identified some 50 patients and are currently collecting information and blood specimens when possible. The study will last at least another year.

In collaboration with Dr. Harry Bartfeld of St. Vincent's Hospital in New York City, lymphocytes from MS patients and controls are being challenged with CNS antigens and tested for their ability to produce migration-inhibitory factors. We have thus far been unable to distinguish between MS and other disease states using this test. It appears however, with the limited number of patients tested that any active brain degeneration, whether it be due to cerebral-vascular accident, metastatic brain cancer, or MS exacerbation can be detected with the test and most probably is non-specific in nature.

We have been attempting to devise in vitro immune assays for measles virus and vaccinia virus, as a result of findings by ourselves and others, that patients suffering from MS possess elevated measles antibody titers and possibly elevated vaccinia antibody titers. We have devised purification procedures for various strains of measles virus and vaccinia virus and have developed an in vitro lymphocyte transformation assay, which we are now using in tests of lymphocytes from MS patients and controls.

Recently Koprowski and his associates reported the isolation of parainfluenza Type I virus from the brain cells of two multiple sclerosis patients. We have completed a serological study using one of these isolates, MS 6/94 as test antigen along with a standard human parainfluenza Type I. We were unable to demonstrate higher HI antibody titers in the sera of MS patients than controls for the MS 6/94 isolate or standard parainfluenza Type I. The etiologic relationship of the virus isolate to MS still remains in question.

In a collaborative study with Dr. James Ganley of the National Eye Institute we tested lymphocytes of patients suffering from presumed ocular histoplasmosis with a large battery of antigens and mitogens. Lymphocytes from patients were more reactive to the mitogens than controls. Moreover, the specific antigens tuberculin and histoplasmin were more stimulatory with lymphocytes of patients than histoplasmin positive controls who did not show the eye manifestation. It appears therefore that individuals with the ocular disease possess a hyperactive cellular immune system.

Studies of ALS and PD

Lymphocyte transformation studies are continuing in our laboratory on Guam using lymphocytes from ALS and PD patients. We are presently receiving shipments of partially extracted lymphocytes from ALS and PD patients that were challenged with CNS antigens. Final extractions and liquid scintillation counting is being performed in our Bethesda laboratory.

Frederick Wolfgram recently has shown that sera from ALS patients in the States and Guam causes lysis of anterior horn cells in culture. We are attempting to set up the technique in our laboratory to further evaluate the specificity of the serum factor and its role in the pathogenesis of ALS. We are particularly concerned with simplifying the system

Selected Studies:

1. An epidemic of diarrhea occurred in Morgantown, Pennsylvania in 1971 affecting at least 60% of the population. Recently, a virus has been isolated from the stool of several cases and identified as a parvovirus. This is the first documented human epidemic with a virus of this group. The parvoviruses in pregnant females of many animal species are associated with teratogenesis and abnormal pregnancies. We therefore checked all pregnancies and deliveries recorded in Morgantown for the past 5 years. We found no evidence of increased abnormal births following the parvovirus epidemic, and hence it does not appear that this parvovirus is associated with severe or obvious birth abnormalities.

2. We are studying the outcome of pregnancy among nurses to determine if those who work in hospital services with high potential contact with virus infections are at increased risk of fetal damage. Of 1125 nurses contacted we have a response rate of about 80 percent (863). Data are being analyzed. To date, among approximately 1800 pregnancies we have about 250 abnormal pregnancies.

II. Laboratory studies related to the epidemiology and immunology of neurologic and perinatal diseases.

Studies on the Minute Virus of Mice (MVM)

MVM was selected as a model agent because of its reported teratogenicity, its propensity for producing chronic infections, and the suggestion that parvoviruses may be important in the etiologies of certain chronic diseases of man and animals. It is our impression that a search for viral nucleic acids, antigens, and/or infectious agents in such diseases would be best approached with methods, reagents and knowledge generated by an indepth study of a typical parvovirus such as MVM.

The development of techniques for cultivation, purification, quantitation and characterization of MVM and its proteins and DNA have continued. Chronic infection *in vitro* has been established with two strains of the virus in mouse L cells while observations on 3 other chronic tissue culture infections have continued.

Further delineation of the secondary structure of MVM DNA appears to confirm our earlier impression that the single-stranded genome contains complementary sequences at two or more different sites, allowing "snap-back" self annealing. Appreciation of this phenomenon is essential to interpretation of data from experiments using DNA-DNA hybridization to demonstrate a parvovirus DNA within cells.

Collaborative studies with Dr. D. Hoggan (LVD, NIAID) have demonstrated that MVM possesses at least 3 proteins. The polyacrylamide gel electrophoretic pattern is similar but distinct from that reported for

other parvoviruses, most closely resembling HADEN (bovine) virus. Differences in the relative concentration of one of the proteins has been observed in preparations cultivated in 2 different cell lines and is currently being investigated.

Recently we demonstrated MVM DNA in both acutely and chronically infected cells in culture as well as in cells from acutely infected newborn mice. This method will be applied to a search for the MVM genome in cells from animals and cultures which appear to have recovered from their primary infection, i.e. for evidence of persistence of the genome in a latent state.

Studies in collaboration with the laboratory of Dr. Samuel Baron, (LVD, NIAID) have shown that MVM is apparently incapable of eliciting an interferon response from infected cells but that, if added to the culture interferon will produce a mild to moderate diminution in MVM replication. This observation is expected to be useful in studies on the termination of chronic MVM infection in vitro and in vivo.

Studies on the viral etiology and molecular pathogenesis of chronic neurologic diseases

Approaches developed through study of the MVM model system are now being applied to a search for virus involvement in multiple sclerosis, amyotrophic lateral sclerosis and Parkinson's disease. Attempts are being made to establish continuous tissue culture from CNS cells in the belief that these might provide a particularly valuable resource. One such line (from the cord of a Guamanian ALS patient, obtained from the Epidemiology Branch Guam laboratory through Dr. C.J. Gibbs) grew slowly but steadily for 4 months, then developed bizarre cytomorphology and the cells died. Supernatent fluids which had been collected periodically are now being pooled and concentrated in preparation for electron-microscopic and biochemical testing. In collaboration with Dr. M. Viola (Head of Medical Oncology, Howard University), CNS tissues from patients with MS, ALS, PD, and SSPE are being tested for the presence of 70S RNA and/or reverse transcriptase activity. As a further extension of these investigations we are preparing reagents (cell DNA, radioisotope tagged DNA "probes") to be used in the search for viral genetic material in DNA extracted from these same tissues. Viral "probes" have been prepared from MVM, measles, vaccinia and Rauscher leukemia viruses; additional probes will be used as they become available. This work represents an application to research on chronic neurologic diseases of ideas and methods developed and used successfully to demonstrate a latent viral genome in human malignancies.

Studies of Multiple Sclerosis

We are continuing in our efforts to define the role of cellular immunity in the pathogenesis of MS. As reported previously, lymphocyte transformation studies involving MS patients, stroke patients and healthy controls have consistently failed to yield significant differences when various CNS antigens were tested.

so that it can be used as a diagnostic field tool for our studies of ALS in Guam and elsewhere.

Studies of SSPE

We have gathered epidemiologic evidence that has led us to postulate that SSPE patients differ from controls in that a large proportion have clinical measles before age one, presumably during the time that some degree of passive immunity exists. We are attempting to reproduce this protection in mice by immunizing the mothers, breeding them and inoculating the passively immune offspring with mouse-adapted measles at various times after birth. The mouse-adapted strain normally kills newborn mice but not adult mice. We have been able to establish passive protection in the newborns as previously reported. We periodically sacrifice representative survivors and search for viral antigens employing cell culture, virologic and fluorescent antibody techniques.

Groups of survivors are being followed which receive injections of live measles-infected vero cells. Other groups received injections of cells obtained by trypsinizing the brains of measles-infected newborns. We have thus far been unable to produce a chronic measles encephalitis in these survivors.

In collaboration with Dr. Michael Viola of Howard University we have preliminary evidence of reverse transcriptase activity in a purified preparation of an SSPE measles strain. A low passage Edmonston strain of measles virus served as control and failed to show any enzymatic activity. We are presently growing up large pools of high titered measles and purifying the preparation to determine the presence of reverse transcriptose activity since this enzyme could be the mechanism whereby measles can become established as a chronic infection in man.

III. The Continuing Activities on Guam and the Trust Territory

In February 1972 a peer review group met on Guam to review the ALS-PD program and make recommendations. They recommended that special emphasis be placed on some specific projects which included several genetic studies, continuing epidemiologic studies of host characteristics and geographic distribution of cases, and neurochemical traits. September 1973 was suggested as a date for further review of the program.

In order to facilitate these recommendations our Deputy Branch Chief, Dr. Dwayne Reed, was sent to Guam for a one year assignment on January 1, 1973. He is currently giving his full attention to completing the epidemiologic and genetic projects.

Computer analysis of descriptive data on more than 560 verified cases of ALS and PD seen at the NINDS Research Center since 1945 have been completed. In agreement with earlier studies this analysis shows that Guam continues to have phenomenally high incidence and mortality rates of ALS and PD, although there has been a slight but consistent decline during

the past 8 years. Cohort analysis indicates that persons born after 1920 have been experiencing a lower risk than persons born earlier.

Three studies of the familial risk of ALS and PD are nearing completion. A family registry of all 0.5 relatives of over 140 cases and an equal number of matched controls originated in 1957, has been updated for new cases and deaths. All offspring of the first 100 cases each of ALS, PD and matched controls, have been followed since 1967. The third study is of the offspring of 27 doubly affected spouse pairs.

Classic genetic studies involving the pedigrees of all 560 verified cases are also being conducted. A subset of these cases, those born in the village of Umatac, have been traced back to 1835 through church and vital statistics records.

Detailed investigation of high and low rate villages has begun and a case-control study of life style and host characteristics is nearing completion.

Present clinical studies are directed towards new case finding, treatment and follow up of known cases. As of February 1, 1973, 109 cases and suspect cases were being followed. Thirty PD cases are currently being treated with L-Dopa MK 486. All have shown remarkable improvement and are being observed for effect of the drugs on life expectancy. The usefulness of L-Dopa and isoprinosine for ALS has been tested for two years.

In cooperation with other researchers we are conducting studies of abnormal dopamine metabolism in the CNS, searching for infectious agents, conducting analysis for environmental toxins, pursuing possible lytic factors for anterior horn cells in serum of ALS patients, searching for immune complexes in kidneys of cases and controls, and mapping amine distribution in brains of affected and controls.

IV. Genetic Studies of Neurologic Diseases

The Genetics Section has continued its interest in disorders of movement, in hereditary nervous system tumors, and in certain hereditary epilepsies.

Observation from sera of 50 individuals with the hereditary torsion dystonias indicate that dopamine-beta-hydroxylase, the enzyme which governs the conversion of dopamine to noradrenalin, is present in abnormal levels in individuals with autosomal dominant torsion dystonia. This work, carried out in collaboration with Dr. Julius Axelrod, may lead to an understanding of the basic biochemical abnormalities in the dystonias, potentially in other movement disorders such as Parkinsonism and Huntington's Chorea. An immediate, practical application is detection of the preclinical state and accurate genetic counseling within families affected by dystonia.

We are nearing completion of a study about Huntington's Disease designed to evaluate the role of a lay organization which provides information and support for affected families. Also surveyed are attitudes of 1,062 Huntington's Disease patients and their relatives towards this tragic disease.

Preliminary analysis indicates that 53 percent of respondents at high risk regarded the lay organization as their best source of information. In contrast, only 15 percent regarded medical specialists such as geneticists as their best source of information with a third group indicating relatives and family physician as their best source. Accurate knowledge regarding risk of transmission was highest in the first group and lowest in the third group. Although respondents in the first group were not as likely to limit family size automatically, they were the most interested in predictive tests on parent or fetus.

One conclusion seems that lay organizations can be a useful, efficient means for the dissemination of information and provision of psychological support for members of families affected with specific genetic disease.

In the area of hereditary eye tumors we are continuing to explore the relationship between hereditary bilateral acoustic neuroma and peripheral neurofibromatosis. Evaluation of melanosomes from café-au-lait spots of patients with bilateral acoustic neuroma is being carried out with the collaboration of the Department of Dermatology, of the Massachusetts General Hospital. If these melanosomes show the same changes as have been described in peripheral neurofibromatosis, it will be strong evidence that both conditions have a common origin. It will also allow a means of detecting carriers of either trait in the presymptomatic period.

Study of retinoblastoma is continuing. We are evaluating plasma dopamine-beta-hydroxylase levels in infants and children with this disorder. This enzyme is elevated in many patients with neuroblastoma, a tumor which shares some histologic features with retinoblastoma.

If the DBH is elevated in those with retinoblastoma, measurement of the enzyme will be a useful screening test to indicate infants at risk as well as a means to evaluate the course of tumor therapy.

The first phase of the progressive myoclonic epilepsy study is nearing completion. 37 affected in 19 families have been ascertained. At least 4 distinct diseases are present within this symptom complex. We are collaborating with several neurochemical laboratories in an attempt to define the biochemical lesion of each.

CONTRACT NARRATIVE
Epidemiology Branch, C&FR, NINDS
Fiscal Year 1973

THE JOHNS HOPKINS UNIVERSITY (NIH71-2026)

Title: The Study of Regional Differences in Stroke Mortality

Contractor's Project Director: Dean M. Nefzger

Current Annual Level: 0 - Ended October 1973

Objectives: The contractor will: (a) Analyze death certificates of all veterans dying in 1967 in Georgia (high mortality area) and five Rocky Mountain States (low mortality area). From the certificates approximately 1,000 certified CVA deaths in each area and 100 randomly selected controls will be chosen for further analysis (2,200 cases total). From these basic data, the frequency of reported CVA among veterans will be compared with male populations in similar areas to determine if the geographic variations reported for civilians occur among veterans; (b) By review of available hospital records and when necessary by physician or family interview, the validity of the diagnosis will be established in order to estimate the relative frequency of mistaken diagnosis or failure to make the diagnosis of CVA; (c) By review of the accumulated information on veterans dying of CVA an estimate of the relative frequency of specific types of CVA will be compiled; (d) All verified stroke deaths and all errors in death certification will be analyzed in terms of geography, age, race, place of residence, marital status, from the point of view of sources of information (competence of certifying individual) and other variables; (e) During this investigation the complete Military and Veterans Administration folders will be reviewed for a subgroup of 50 cases and controls per state in order to evaluate the usefulness of these records in subsequent studies. In addition, all other avenues of ascertainment of a valid rate of CVA among veterans will be explored in order that a definitive study of veterans population be conducted in the future when the great bulk of veterans of the Second World War arrive at the age of high risk for CVA.

Major Findings: A total of approximately 1,000 stroke deaths and 2,800 deaths from other causes have been compiled. The data on these cases were analyzed. A preliminary review of the stroke cases in Georgia which has a high mortality rate from stroke versus the Rocky Mountain States which have a low reported mortality from strokes has revealed surprising and potentially important information. The updated results indicate a pattern of greater risk in Georgia than the Mountain States thus supported previous observations. However, in contrast to other studies the difference was entirely due to an excess of deaths in Georgia from hemorrhagic stroke. Ischemic stroke rates were equal in both areas. The data were not changed when race was controlled.

Significance to NINDS Program and Biomedical Research: The epidemiological patterns of stroke are poorly understood, although it is suspected that there are regional differences throughout the U.S. Any information confirming these differences and indicating a cause for these differences could lead to a better understanding of causation and prevention in this important cause of morbidity and mortality.

Proposed Course of Project: The contract expired, but final processing of data is not complete. The investigators received a contract of $21,937 to complete this work. We are awaiting a final report. Also, this study population is being reviewed with the prospect of early (preinduction) risk factors which might have been determinants in future development of stroke.

Publications:

Acheson, R.M., Nefzger, M.D. and Heyman, A.: Mortality from Stroke among U.S. Veterans in Georgia and Five Western States. V. Seventh and Eighth Revisions of the International Classification of Diseases as They Relate to Stroke. Amer. J. Epid. 96:396-400, 1973.

Nefzger, M.D., Acheson, R.M. and Heyman, A.: Mortality from Stroke among U.S. Veterans in Georgia and Five Western States. I. Study Plan and Death Rates. J. Chron. Dis. In press.

Acheson, R.M., Nefzger, M.D. and Heyman, A.: Mortality from Stroke among U.S. Veterans in Georgia and Five Western States. II. Quality of Death Certification and Clinical Records. J. Chron. Dis. In press.

Acheson, R.M., Nefzger, M.D., and Heyman, A.: Mortality from Stroke among U.S. Veterans in Georgia and Five Western States. III. Hypertension and Demographic Characteristics. J. Chron. Dis. In press.

Heyman, A., Nefzger, M.D. and Acheson, R.M.: Mortality from Stroke among U.S. Veterans in Georgia and Five Western States. IV. Clinical Observations. J. Chron. Dis. In press.

Nefzger, M.D., Acheson, R.M. and Heyman, A.: Mortality from Stroke among U.S. Veterans in Georgia and Five Western States. VI. Clinical Validation of Death Certificates. J. Chron. Dis. In press.

CONTRACT NARRATIVE
Epidemiology Branch, C&FR, NINDS
Fiscal Year 1973

NATIONAL RESEARCH COUNCIL, FOLLOW-UP AGENCY (PH43-64-44 [Task Order 53])

Title: New Epidemiologic Study of Multiple Sclerosis in U.S. Military Veteran Population

Contractor's Project Director: Gilbert Beebe, M.D.
James Norman, M.D.
John F. Kurtzke, M.D.

Current Annual Level: $34,700

Objectives: The contractor will perform an extensive survey of multiple sclerosis among veterans of the Second World War and the Korean War. This will be an update of an initial survey which included 600 patients and will include approximately 6,000 patients. Patients will be matched with controls to determine geographic patterns, socio-economic status, urban-rural localization and numerous other variables which have been tested for multiple sclerosis. Parallel information will be available for Negro MS patients and controls and white female MS patients and controls. In addition an extensive investigation of migrant and non-migrant populations to and from high and low risk areas for multiple sclerosis will be conducted. Emphasis will also be placed on the relationship between the communicable diseases experienced in childhood and subsequent multiple sclerosis.

Major Findings: Data collection is proceeding. Material will be analyzed during the third year of this contract. Considerable effort is now being expended in identification and controlling of Korean War veterans.

Significance to NINDS Program and Biomedical Research: Multiple sclerosis is a major neurologic disease in the United States. Previous epidemiologic studies have indicated that there is in the Northern Hemisphere a north-south gradient in the rates of multiple sclerosis. From a previous study, certain interesting correlations were noted among military personnel who developed multiple sclerosis, such as an excess of people with higher I.Q.s, with urban residence, with higher socioeconomic status and with defective eyes. The recent emphasis on risk of multiple sclerosis among migratory populations is of crucial importance since it must be determined if the causative factors of this disease are more prevalent in northern areas or if there are protective factors or mechanisms operating in southern areas. If clear patterns can be delineated by this study, the results could suggest in which populations we must concentrate in the search for the etiology and prevention of multiple sclerosis.

Proposed Course of Project: This project was initiated in June 1972 and will run for three years. The total 3-year cost is $144,523.

NATIONAL ACADEMY OF SCIENCES (PH43-64-44 [Task Order 62])

Title: Epidemiologic and Follow-up Study of Amyotrophic Lateral Sclerosis and Other Motor Neuron Diseases

Contractor's Project Director: James Norman, M.D.
Gilbert Beebe, M.D.
John F. Kurtzke, M.D.

Current Annual Level: $35,848.00

Objectives: 1. An epidemiologic evaluation of potential risk factors among approximately 500 patients with various forms of motor neuron diseases will be conducted. All deaths among U.S. males coded 365.1 (ALS) for 1963-1967 will be collected and those who are veterans will be selected for the study. Controls will be based on National Service Life Insurance holders in a fashion similar to the ongoing multiple sclerosis veterans study. Pertinent data will be collected from various induction and military service records and coded, and significant differences between patients and controls will be sought. A subsample of patients records will be reviewed to confirm the death certificate diagnosis of ALS. 2. A study of the natural history of the various motor neuron diseases including ALS will be investigated from Veterans Hospital admissions from 1957 to 1964, who were diagnosed as having motor neuron disease. The total number should be approximately 500 of whom approximately 200 should have motor neuron diseases other than ALS. The epidemiologic patterns as well as the clinical course of the various entities now grouped under motor neuron disease will be analyzed and compared to see if distinctive patterns emerge. In addition, through this technique the first life table of ALS will be developed by using a stratified age of onset sampling method. 3. A detailed case-control study will be conducted involving 200 living ALS patients and matched controls. With this mechanism more specific information on life styles and patterns of residence, employment, etc. can be analyzed which would not be available through abstracting military induction of Veterans Administration Hospital records. The 200 patients and 200 controls will be selected from the same VA Hospital Service and interviewed by trained social workers in the VA System. Since no true leads to the etiology of ALS are known, the questionnaire is extensive but has been shown to be workable in field tests of some 30 patients and equal number of controls.

Major Findings: Data collection for the three phases of this project have proceeded during the first year. Approximately 50 questionnaires have been received and checked for completeness for Part 3 (see above).

Significance to NINDS Program and Biomedical Research: Regional differences and socioeconomic differences have been commented upon

for amyotrophic lateral sclerosis in the United States but definitive data are lacking. This study will determine for a large population if there are predisposing factors to this disease. Should we determine such factors, it would provide a valuable clue to the understanding, treatment and prevention of this disease. Prognosis for survival should also be clarified.

Proposed Course of Project: This project is to run three years. Budget for the second year will be $43,996.

CONTRACT NARRATIVE
Epidemiology Branch, C&FR, NINDS
Fiscal Year 1973

KAISER FOUNDATION RESEARCH INSTITUTE (NINDS72-2319)

Title: Risk Factors for Stroke

Contractor's Project Director: G. Browne Goode, M.D.
G. Friedman, M.D.

Current Annual Level: Project to run from June 30, 1972 - December 29, 1973 at a total cost of $91,440

Objectives: 1. Identify patients admitted to the San Francisco Kaiser Foundation Hospital and Outpatient Clinic, with the diagnosis of CVD, during the period 1965-1971, who received a multiphasic screening exam prior to onset of the stroke. 2. Review the patient charts for validation of the diagnosis and grouping of the patients into various categories of CVD. 3. Select two types of control patients without evidence of CVD; one group matched to the cases by known risk factors of CVD who had received the multiphasic screening exam, and also the entire population which received multiphasic screening stratified by age and sex. 4. Obtain by computer search and transfer to a separate computer tape the stored multiphasic examination data for the case group and two control groups. 5. Conduct statistical comparisons of the case group with the two control groups for each variable and measurement included in the multiphasic examination. Statistical tests of differences will be used for individual items, and more complex statistical analysis will be used to determine the interaction of variables which differentiate the cases from the controls.

Major Findings: In the first six months of this project, the contractors have identified and confirmed all cases of cerebrovascular disease (CVA) with prior multiphasic check-ups (MHC), who were admitted to Kaiser Foundation Hospital, San Francisco from 1965 to 1971. Approximately 400 cases have been identified. They have devised a set of definitions of what constitutes a CVA, a transient ischemic attack (TIA), and have devised a classification of CVA's. A 6 page coding form for hospital record review, containing digital data for future use by keypunch operators, has been revised several times and is now in a final form. To date, approximately half of the 400 cases have been reviewed sequentially by the research medical librarian and then the principle investigator. Some cases have additionally been reviewed by the co-investigator.

Significance to NINDS Program and Biomedical Research: Stroke is the fourth cause of death in the United States. By studying a well-defined population who received prior multiphasic screening we will increase knowledge on the relative affects of various risk factors and through using controls subtle and previously unrecognized risk factors may emerge.

Proposed Course of Project: In the next 6 months, we anticipate finishing the subject cases chart reviews, and then we will move on to review some outpatient CVA patients with prior MHC. The two control groups will be obtained after the current aspect of this study is completed.

CONTRACT NARRATIVE
Epidemiology Branch, C&FR, NINDS
Fiscal Year 1973

THE UNIVERSITY OF TEXAS AT HOUSTON (NO1-NS-3-2304)

Title: Epidemiological Studies of Stroke Risk Factors in Panama

Contractor's Project Director: Stephen Bennett, M.D.
Reuel Stallones, M.D.
Darwin Labarthe, M.D.

Current Annual Level: $118,857.00 (Feb. 28, 1973 - Feb. 27, 1974)

Objectives: To design and carry out an epidemiologic survey of certain populations in Panama, to study the association of genetic and environmental factors with cerebrovascular risk. At the conclusion of the field survey, the data will be analyzed by computer and will consist of descriptive tabulations of study variables by appropriate subgroupings, and the association of genetic and environmental factors with cerebrovascular risk factors, both among and within the population subgroups.

Major Findings: The feasibility and organizational aspects have been worked out. Populations and schedules were developed. The framework for a subcontract with the Faculty of Medicine of the University of Panama has been developed.

Significance to NINDS Program and Biomedical Research: Rates of hypertension and stroke are approximately twice as high among blacks as among whites in the United States. There is, however, little information related to the association of these risk factors to different life styles, and genetic influence. This cross-sectional survey would yield valuable information concerning the comparative prevalence of CVD precursor disease states among defined black populations with different origins and of different socioeconomic levels. Such contrast could shed light upon the relative importance of the genetic versus environmental factors.

Proposed Course of Project: This project will run approximately 2 years. The second year's budget is approximately the same as the first year ($19,000).

Serial No. NDS (CF) - 55 E 201
1. Collaborative & Field Research
2. Epidemiology Branch
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Studies on amyotrophic lateral sclerosis/parkinsonism-dementia complex of Guam (ALS-PD)

Previous Serial Number: Same

Principal Investigators: Jacob A. Brody, M.D.
Dwayne Reed, M.D.
E. Michael Holden, M.D.
NINDS Research Center

Other Investigators: Jose Torres
NINDS Research Center
Francisco Leon Guerrero
NINDS Research Center
Manuel T. Cruz
NINDS Research Center
Olivia Cruz, M.D.
NINDS Research Center
Thomas N. Chase, M.D.
NIMH

Consultants: Kwang-Ming Chen, M.D.
Guam Memorial Hospital
Yoshiro Yase, M.D.
Wakayama Medical College, Japan
Leonard T. Kurland, M.D.
The Mayo Clinic, Rochester, Minnesota
Donald W. Mulder, M.D.
The Mayo Clinic, Rochester, Minnesota
Haruo Okazaki, M.D.
The Mayo Clinic, Rochester, Minnesota

Cooperating Units: NINDS Research Center, Tamuning, Guam
Special Chronic Disease Studies, C&FR, NINDS
Laboratory of Slow, Latent, and Temperate Viruses, C&FR, NINDS
The Mayo Clinic, Rochester, Minnesota
Department of Pathology, Massachusetts General Hospital, Boston
Amino Acid Laboratory, Massachusetts General Hospital, Boston
Department of Neurology, Wakayama Medical College, Wakayama, Japan

Medical Research Center, Brookhaven National Laboratory, Upton, New York (George C. Cotzias, M.D.)
Trust Territory Health Office
Division of Neurology, Center of Health Sciences, University of California at Los Angeles (Frederick Wolfgram, M.D.)
Laboratory of Clinical Sciences, NIMH
School of Public Health, University of Hawaii, Honolulu
Unit on Neurology, NIMH
Department of Neurobiology, Clinical Research, Institute of Montreal (Andre Barbeau, M.D.)
Neuropathology Branch, Armed Forces Institute of Pathology, Washington, D.C.

Man Years:

Total:	1 1/2
Professional:	1/2
Other:	1

Project Description:

Objectives: To determine the cause of ALS and PD, and to determine the epidemiological, clinical, neuropathological and physiological significance of these diseases and to develop therapeutic approaches to the diseases.

Methods employed: Routine methods for epidemiological, clinical, neuropathological, and neurochemical and therapeutic investigations.

Major findings: In February 1972 a peer review group met on Guam to review the program and to make recommendations. This review was extremely valuable to us as it served to develop an overall perspective to the program and to point out some sharply defined targets for our research focus. The general recommendation was that the program continue pursuing the clinical, epidemiologic, genetic, and collaborative studies with special emphasis upon the following: 1) completion of the registry and pedigree genetic study described elsewhere [NDS (CF) - 67 E 1487], 2) mortality and demographic analysis to facilitate cohort studies, life table analyses, complete ascertainments of deaths, and fertility of cases and controls, 3) case-control epidemiologic surveys of the living cases, to search for environmental and life-style differences among affected and unaffected persons on Guam, 4) immigrant and emigrant studies to more clearly establish the risk of ALS and PD among both Chamorro groups living on islands other than Guam and non-Chamorro persons living on Guam, 5) completion of the therapeutic trials and neurochemistry studies.

In order to facilitate this program, the Deputy Branch Chief, Dr. Dwayne Reed, was assigned to Guam for one year, beginning January 1, 1973. While most of these projects are continuing, some preliminary findings are noted below.

As of February 1, 1973 we were following a total of 108 patients; including 46 confirmed ALS, 3 suspect ALS, 40 confirmed PD and 11 suspect PD. During the calendar year 1972 there were 7 deaths from ALS, and 8 deaths from PD.

Analysis of the temporal and geographic patterns of ALS and PD on Guam from 1950-1972 indicate that since 1965 there has been a gradual but definite decrease in both incidence and mortality rates. This is the first time such a change has been noted. Within the confines of Guam, the southern villages have had strikingly higher rates than other villages. It is in these same southern villages that the most dramatic rate changes have taken place. We are currently trying to discover environmental changes which may be associated with these changes.

We are also following and reviewing the occurrence of ALS and PD among Chamorros living on other islands of Micronesia. Preliminary findings indicate that they have the same high rate as Guam.

Case-control studies of host characteristics and studies of high and low rate village environments are underway. Major emphasis has been placed upon examinations of soil, food and water, and human tissue samples for the presence of toxic heavy metals.

Present clinical studies are directed towards new case finding and follow-up and treatment of active cases. Double-blind studies of isoprinosine and L-Dopa on ALS patients have just been completed and the analysis will be finished in the near future. Preliminary examination indicates no difference between the treatment and placebo groups. Thirty PD patients are being treated with L-Dopa - MK486 and we are observing them closely to determine if this treatment extends their life expectancy. All have significant improvement in their extrapyramidal function.

In cooperation with Dr. Thomas Chase, Chief, Section on Experimental Therapeutics, Laboratory of Clinical Sciences, NIMH, we have initiated a series of studies to determine abnormal dopamine metabolism in the CNS utilizing expired air and urine. This is a most important aspect of our future work as, if successful, it will permit us to conduct population surveys of abnormal dopamine metabolism which are now impossible to do because of the necessity to use spinal fluid. In addition, we have completed analysis of secondary movement disorders among PD and ALS patients receiving L-Dopa. While these abnormalities were noted in more than one-half of PD patients they did not appear in any of 14 treated ALS patients.

Neuropathologic descriptions of the ALS and PD brains continue to point out the wide distribution of neurofibrillary degeneration. In addition, studies of 46 control brains from Chamorros who died of causes other than ALS or PD indicate that over two-thirds of adult Chamorros have neurofibrillary degeneration in their central nervous system. Continued efforts are being made to collect as many control brains as possible from Chamorro, Filipino, and other island residents.

A number of collaborative studies are in process utilizing brain tissue and blood from the Guam cases. These projects include the search for a causative virus, studies of environmental toxins, and the search for immune complexes in the kidneys of cases and controls. Recently Dr. Frederick Wolfgram in a "blind" series of ALS and PD patients and controls was able to detect a serum lytic factor in 4 of 5 ALS patients that did not occur in PD patients or controls. Dr. Andre Barbeau has done amine mapping of the cerebrum of PD and ALS patients and controls. Preliminary results indicate that cellular dopamine was greatly depressed in PD patients but not in LAS patients or controls. It is difficult to interpret this finding in view of the observed decreased CNS dopamine metabolism in ALS patients as well as in PD patients.

Significance to biomedical research and the program of the Institute: Guam has the highest incidence in the world of motor neuron disease and the unique disease PD. The documentation of the epidemiological, clinical, and neuropathological aspects of ALS and PD have added to the world's knowledge concerning these neurologic diseases. In fields in which there are no known causes and no known cures, data such as these provide one of the most likely avenues for development of concepts and facts which lead to causes and cures. We are also exploiting this unique opportunity to test new drugs of potential benefit to patients. Through our studies we have discovered a dopamine deficiency in ALS patients on Guam and in the U.S. that is the first promising lead in the understanding of this disease.

Proposed Course: Our major efforts are to complete the current projects by October 1973 at which time a second peer review group will meet on Guam.

Honors and Awards: None

Publications:

Reed, D. and Brody, J.A.: Amyotrophic lateral sclerosis and parkinsonism-dementia of Guam; recent observations. Presented at the annual Society for Epidemiologic Research in Houston, Texas, June 1972.

Reed, D., Labarthe, D. and Stallones, R.: Epidemiologic studies of serum uric acid levels among Micronesians. Arthritis Rheum. 15:381-390, 1972.

Stanhope, J.M., Brody, J.A. and Morris, C.E.: Epidemiologic features of amyotrophic lateral sclerosis and parkinsonism-dementia in Guam, Mariana Islands. Int. J. Epid. 1:199-210, 1972.

Reed, D., Labarthe, D., Stallones, R. and Brody, J.A.: Epidemiologic studies of serum glucose levels among Micronesians. Diabetes, 22:129-136, 1973.

Brody, J.A. and Kurland, L.T.: Amyotrophic lateral sclerosis and parkinsonism-dementia in Guam. In Tropical Neurology, Oxford University Press. In press.

Brody, J.A., Stanhope, J.M. and Kurland, L.T.: Patterns of amyotrophic lateral sclerosis and parkinsonism-dementia of Guam. In Contemporary Trends: Neurological Problems in Oceania. Editor: R.W. Hornabrook, New Guinea. In press.

Kurland, L.T. and Brody, J.A.: Amyotrophic lateral sclerosis with special reference to the Guam type. In Handbook of Clinical Neurology. Editors: P.J. Vinken and G.W. Bruyn, Amsterdam. In press.

Shih, V.E., Brink, E.W., Peneva, P. and Brody, J.A.: Blood and urinary amino acid patterns in Guamanians and Micronesians. In press.

Serial No. NDS (CF) - 63 E 1103
1. Collaborative & Field Research
2. Epidemiology Branch
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Neurological diseases other than ALS/PD on Guam

Previous Serial Number: Same

Principal Investigator: E. Michael Holden, M.D.
NINDS Research Center

Other Investigators: Kwang-Ming Chen, M.D.
Guam Memorial Hospital
Jacob A. Brody, M.D.
Leonard T. Kurland, M.D.
Mayo Clinic
Dwayne Reed, M.D.

Cooperating Units: NINDS Research Center, Tamuning, Guam
Mayo Clinic, Rochester, Minnesota

Man Years:

Total:	9/16
Professional:	1/16
Other:	1/2

Project Description:

Objectives: A survey in 1954 by Donald W. Mulder, M.D. and Leonard T. Kurland, M.D. gave the impression that not only ALS, but also other heredofamilial neurologic disorders seemed unusually prevalent while multiple sclerosis and perhaps CNS tumors are uncommon. The objective of this study is to try to determine the validity of this data and to see if it is related to ALS and PD.

Methods employed: Between 1956 and the end of 1971, the NINDS Research Center occupied a nearly unique position on Guam. It was the only neurological consultation service available to all ethnic groups on Guam and saw most neurological patients at Guam Memorial Hospital and Naval Hospital. Since then, with the advent of Dr. Kwang-Ming Chen, to the island, as a permanent resident, and with liberalization of hospital privileges of the native population regarding access to the Naval Hospital, there have been three neurological consultation services available to native-born Guamanians. It is still true that virtually all of the patients with ALS or PD are eventually referred to the NINDS Research Center. Similarly, individuals who are suspected of having these two diseases are also referred in large numbers to

this research center. Patients with other neurological disorders however, are being seen by the three consultation services noted above.

Major findings: During the year 120 patients with neurologic diseases other than ALS and PD were seen and 100 EEG's were performed. From 1960 through 1966, in conjunction with ongoing studies of amyotrophic lateral sclerosis and parkinsonism-dementia on Guam, 1,028 Chamorro patients were referred to our neurologic clinic. In comparison with other populations and particularly that of Rochester, Minnesota, the residents of Guam had higher rates of convulsive disorders, myotonic dystrophy, peroneal muscular atrophy, and hereditary ataxias. There was no indication of an unusual incidence of central nervous system neoplasms, and no cases of progressive muscular dystrophy, myasthenia gravis, or indigenous multiple sclerosis were seen. No patient with proved classic paralysis agitans was observed in the Chamorro population.

Significance to biomedical research and the program of the Institute: This study adds to the general body of knowledge being collected by the Branch regarding the island of Guam and provides information on diseases possibly related to ALS and PD.

Proposed course: We are expanding studies of neurologic diseases on Guam and the Trust Territories using the same techniques.

Honors and Awards: None

Publications: None

Serial No. NDS (CF) - 66 E 1319
1. Collaborative & Field Research
2. Epidemiology Branch
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: A search for autoimmune mechanisms in the pathogenesis of chronic neurological diseases by the use of peripheral lymphocytes

Previous Serial Number: Same

Principal Investigators: Jacob A. Brody, M.D.
George Nemo, Ph.D.

Other Investigators: Harry Bartfeld, M.D.
New York University

Cooperating Unit: Department of Medicine, New York University

Man Years:

Total:	1/8
Professional:	1/16
Other:	1/16

Project Description:

Objectives: To study the role of cellular immunity in the pathogenesis of neurologic disorders suspected to be of autoimmune etiology.

Methods employed: Peripheral lymphocytes from patients with multiple sclerosis (MS) were challenged in vitro with specific antigens. The incorporation of tritiated thymidine into DNA during the synthetic phase of lymphoblast transformation is used as an indicator of lymphocyte responsiveness and is measured using liquid scintillation spectrometry.

An alternative cellular immune procedure is currently being explored. Lymphocytes from MS patients and controls are challenged with brain antigens and tested for their ability to produce migration-inhibitory factor (MIF). MIF is a pharmacologically-active substance elaborated by sensitized lymphocytes in response to a specific antigen. It is characterized primarily by its ability to prevent the migration of macrophages from the open end of a capillary tube.

Major findings: As reported previously, lymphocyte transformation studies involving MS patients, stroke patients and healthy controls have consistently failed to yield significant differences when various brain antigens such as MS brain, normal brain and basic patterns were tested

in our laboratory and in most other laboratories. Positive results have been reported, however, for the MIF.

Of 19 patients and controls thus far tested using the MIF test, 5 of 9 MS patients were positive and 5 of 10 controls were positive. Of the 5 positive patients all 5 were in the exacerbated stage of the disease. Of the 5 positive controls 3 were recovering from recent strokes and 2 had metastatic brain cancer.

It appears with the limited number of patients tested, brain degeneration whether it be CVA, cancer or MS exacerbation can be detected with the test and is most probably nonspecific in nature.

Significance to biomedical research and the program of the Institute: Since it is well established that the lymphocyte is the mediator of cellular immunity, the lack of a significant lymphocyte response as demonstrated in our study casts serious doubt on the hypothesis that MS is an autoimmune disorder.

Proposed course: Macrophage-migration inhibition tests upon MS patients and controls are continuing.

Honors and Awards: None

Publications: Brody, J.A., Harlem, M.M., Plank, C.R. and White, L.R.: Freezing human peripheral lymphocytes and a technique for culture in monolayers. Proc. Soc. Exp. Biol. & Med. 129: 968-972, 1968.

Brody, J.A., Harlem, M.M., Kurtzke, J.F. and White, L.R.: Unsuccessful attempt to induce transformation by cerebrospinal fluid in cultured lymphocytes from multiple sclerosis patients. New Eng. J. Med. 279:202-204, 1968.

Serial No. NDS (CF) - 66 E 1320
1. Collaborative & Field Research
2. Epidemiology Branch
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Stateside Guamanian study

Previous Serial Number: Same

Principal Investigator: Jacob A. Brody, M.D.

Other Investigator: None

Cooperating Units: NINDS Research Center, Tamuning, Guam
School of Public Health, University of California, Berkeley

Man Years:

Total:	3/16
Professional:	1/16
Other:	1/8

Project Description:

Objectives: This study was instituted in July 1966 to determine if ALS and PD occur with the same high frequency among Guamanians who have left Guam. Since the bulk of the stateside Guamanians are in California, efforts have been concentrated there.

Methods employed: A household census was completed in the fall of 1967 which included information on neurologic disease. Names of heads of households were obtained from relatives on Guam, the office of the Guamanian representative to Congress, local Guamanians, social organizations, an 8 year old NINDS California Guamanian registry and from other Guamanians already living in this country. Household information was obtained by trained Guamanian interviewers living in California and by personnel from the Branch. Follow-up examination of suspect cases of ALS and PD was conducted by specialist physicians.

Major findings: The ALS rate in California is as high as it is on Guam. Two patients with presumed Parkinson's disease and/or dementia have died and autopsy studies in one are consistent with parkinsonism-dementia although the patient's age is unusually advanced and in other changes were more compatible with paralysis agitans without dementia as seen in the United States. These findings were included in a report by Brody and Hirano.

Significance to biomedical research and the program of the Institute: The results suggest that a genetic factor and/or early exposure to an environmental factor is responsible for ALS and PD on Guam. The PD patient in California is the only PD patient ever encountered off Guam and the Marianas. The disease is not known to occur in non-Chamorros. Since the rate of PD off Guam is lower than that of ALS it suggests that these diseases are not a spectrum of CNS diseases with a single cause. The patient with paralysis agitans is the first documentation of this disease in a Chamorro. He lived in the United States for 8 years which may be a clue as to the incubation period of paralysis agitans.

Proposed course: While it is planned to maintain contact with this migrant population over the years because of the valuable clues we may gain regarding etiology of amyotrophic lateral sclerosis and parkinsonism-dementia as seen on Guam, this project is completed.

Honors and Awards: None

Publications:

Eldridge, R., Rosario, J. and Brody, J.A.: Amyotrophic lateral sclerosis and parkinsonism-dementia in a migrant population from Guam. (A preliminary report.) In Trans. Amer. Neurol. Ass. 93:204-206, 1968.

Eldridge, R., Ryan, E., Rosario, J. and Brody, J.A.: Amyotrophic lateral sclerosis and parkinsonism-dementia in a migrant population from Guam. (A full report.) Neurology, 19:1029-1037, 1969.

Reed, D., LaBarthe, D. and Stallones, R.: Health effects of westernization and migration among Chamorros. Amer. J. Epid. 92:94-112, 1970.

Hankin, J., Reed, D., Labarthe, D., Nichaman, M. and Stallones, R.: Dietary and disease patterns among Micronesians. Amer. J. Clin. Nutr. 23:346-357, 1970.

Reed, D., Labarthe, D. and Stallones, R.: Epidemiologic studies of serum uric acid levels among Micronesians. Arthritis Rheum. 15:381-390, 1972.

Reed, D., Labarthe, D., Stallones, R. and Brody, J.: Epidemiologic studies of serum glucose levels among Micronesians. Diabetes, 22:129-136, 1973.

Serial No. NDS (CF) - 66 E 1321
1. Collaborative & Field Research
2. Epidemiology Branch
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Japanese encephalitis on Guam

Previous Serial Number: Same

Principal Investigators: Roger Detels, M.D.
Jacob A. Brody, M.D.
C. Joseph Gibbs, Ph.D.
Laboratory of Slow, Latent and Temperate Viruses, NINDS

Other Investigators: None

Cooperating Unit: Laboratory of Slow, Latent and Temperate Viruses, NINDS

Man Years:

Total:	9/16
Professional:	1/16
Other:	1/2

Project Description:

Objectives: A Japanese encephalitis epidemic occurred in 1947 on Guam, but, according to serologic studies, involved only 20% of the population before apparently disappearing from the island. It is the objective of this study to determine if Japanese encephalitis virus (JEV) is persisting on Guam at a low level and to determine why it has not established an epidemic pattern as in Japan, Taiwan and Korea or an endemic pattern as in Malaysia, despite the presence of a suitable vector and reservoir hosts.

Methods employed: Sera will be collected from Guamanians born prior to, during and after the occurrence of the 1947 epidemic and will be analyzed for antibody to JEV and other Group B and Group A arboviruses. Sera will also be collected for antibody screening from animals. Mosquitoes will be collected to determine the types of culicenes present on the island which might act as vectors.

Major findings: Eighteen percent of sera from 498 Guamanians born since 1900 contain hemagglutination inhibition antibodies to JEV. Twenty-one percent born prior to 1950 and 8% born since 1950 have HI antibody to JEV suggesting that there has been Group B arbovirus activity on Guam since 1950. Nonetheless, only 1 of 100 pigs bled had HI antibody to JEV. Culex tritaeniorhynchus, but not Culex annulus, has been identified on the island.

Significance to biomedical research and the program of the Institute: JEV was thought to have disappeared from Guam contrary to the usual pattern. However, the finding of HI antibodies to JEV in 8% of Guamanians born since 1950 but not in pigs when all the known necessary ingredients are present for JEV to be either endemic or epidemic invite investigation to determine the uniqueness of Guam and thus contribute to the knowledge of factors important to the epidemiology of JEV.

Proposed course: This project has been terminated.

Honors and Awards: None

Publications: None

Serial No. NDS (CF) - 67 E 1485
1. Collaborative & Field Research
2. Epidemiology Branch
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Analyses of abnormal urine and blood amino acids metabolism among Guamanians

Previous Serial Number: Same

Principal Investigators: Jacob A. Brody, M.D.
Vivian Shih, M.D.
Massachusetts General Hospital

Other Investigators: Jose M. Torres
NINDS Research Center
Manuel T. Cruz
NINDS Research Center

Cooperating Units: NINDS Research Center, Tamuning, Guam
Amino Acid Laboratory, Massachusetts General Hospital, Boston, Massachusetts

Man Years:

Total:	9/16
Professional:	1/16
Other:	1/2

Project Description:

Objectives: Earlier observation indicated that indigenous Guamanians have difficulties in handling protein and carbohydrate. However, the relationship between the observed hyperuricemia and hyperglycemia on Guam and the neurologic manifestations is not clear.

Methods employed: A contract with the Amino Acid Laboratory of the Massachusetts General Hospital for the broad testing for inborn errors of metabolism was secured and blood and urine are being sent to the lab from Guam.

Major findings: No major abnormalities in infants, retarded children and ALS or PD patients were encountered in the survey of blood and urine in 435 patients with various diseases and 574 normal infants and 20 normal adults in Guam and other islands of Micronesia. One girl apparently healthy but with a history of seizure disorder (apparently a febrile convulsion) many years ago was found to excrete an unknown sulfur amino acid. We are reviewing her family clinically and collecting appropriate urine samples

for confirmation of this finding. No specific changes were found in PD or ALS patients. B-amino isobutyric aciduria was detected in 56.8% of normal infants. Taurine excretion was prevalent in normal infants on the Caroline Islands; it was probably related to breast-feeding. Cystathioninuria was present in 9 normal infants. We have the impression that the infant population in Guam and other areas have relatively low rates of abnormal amino acids when compared with other populations. This cannot be claimed conclusively because sampling methods differed in our series. The Department of Public Health of the Government of Guam has routinely screened all new borns for PKU for the past 6 years and no case has been encountered.

Significance to biomedical research and the program of the Institute: This study contributed to knowledge of metabolic abnormalities of the Chamorro people of Guam.

Proposed course: This project has been terminated.

Honors and Awards: None

Publications: Shih, V.E., Brink, E.W., Peneva, P. and Brody, J.A.: Blood and urinary amino acid patterns in Guamanians and Micronesians. In preparation.

Serial No. NDS (CF) - 67 E 1486
1. Collaborative & Field Research
2. Epidemiology Branch
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1971 through June 30, 1972

Project Title: Torsion Dystonia - a clinical and genetic study

Previous Serial Number: Same

Principal Investigator: Roswell Eldridge, M.D.

Other Investigators: Irving S. Cooper, M. D.
St. Barnabas Hospital
Morris B. Gross, M. D.
Hunter College in the Bronx
Wolfgang Zeman, M. D.
University of Indiana
Mary Coleman, M. D.
Children's Hospital
Frederick Wooten, M. D.

Cooperating Units: Department of Neurologic Surgery, St. Barnabas Hospital, New York
Laboratory of Clinical Science, NIMH

Man Years:

Total:	3/8
Professional:	1/4
Other:	1/8

Project Description:

Objectives: Torsion dystonia (TD) comprises a heterogeneous group of conditions characterized by disordered movement. TD may be due to either genetic or environmental factors. The present study has already largely defined the nosology of these conditions. Further clinical and biochemical family study may suggest the basic defect in each.

Methods employed: Initially, probands with a history of TD selected through 180 neurologic and neurosurgical centers provided the families for study. Recently, physicians and affected individuals themselves have contacted us requesting help. A detailed clinical family history is obtained. The latter stresses geographical origin of ancestral couples. The proband and all available relatives were given physical examinations. Patients from all areas of the U. S. treated by various methods are seen to avoid geographic, ethnic, and therapeutic bias.

Major findings: Clinical, genetic, psychometric and therapeutic aspects of dystonia have been evaluated in more than 200 patients in 130 families. The results have appeared in publications indicated below. Among the conclusions are: at least two hereditary forms of dystonia exist which frequently can be distinguished on clinical grounds; psychotherapy has a limited role as primary treatment; drugs reported to be helpful in the dystonias generally have been ineffective in most patients over a long period; and recent neurosurgical procedures offer hope.

Recently, observations in 20 individuals from four families with torsion dystonia have suggested that the enzyme dopamine betadehydroxylase, (DBH), which governs the conversion of dopamine to noradrenamine, is present in abnormal levels in individuals with the dominant form of torsion dystonia. This work is being carried out in the laboratory of Dr. Julius Axelrod.

Significance to biomedical research and the program of the Institute: Elucidation of the fundamental defect in these forms of dystonia will be of practical importance. In addition to suggesting specific treatment, it should be possible to distinguish between the recessive and dominant forms chemically. The application to genetic counselling of such a test is obvious. As in other inborn errors of metabolism, such a study could provide basic, new information about central nervous system physiology.

Parkinson's disease shares certain clinical features with dystonia and is relieved by the same operative procedure so that information gained from the dystonia study may bear on this important problem.

Proposed course: This Project is Terminated.

Honors and Awards: None

Publications: Eldridge, R., Ryan, E., Brody, J.A. and Cooper, I.S.: Dystonia musculorum deformans: Evidence for two hereditary forms. Excerpta Medica International Congress Series No. 175, Progress in Neuro-Genetics, Vol. I of the Proceedings of the Second International Congress of Neuro-Genetics and Neuro-Ophthalmology, Montreal, September 1967, pp. 772-788, 1969.

Eldridge, R., Harlan, A., Cooper, I.S. and Riklan, M.: The Hereditary Torsion Dystonias (Dystonia Musculorum Deformans): Geographical distribution and I.Q. in dominant and recessive forms. In Transactions of the American Neurological Association, 94, 1969.

Eldridge, R., Harlan, A., Cooper, I. S. and Riklan, M.: Superior intelligence in recessively inherited torsion dystonia. The Lancet I:7637, pp. 65-67, 1970.

Eldridge, R., Edgar, A., and Cooper, I.S.: Genetics, Geography and Intelligence in the torsion dystonias. Proceedings of the Second Conference on the Clinical Delineation of Birth Defects, May 1969. The National Foundation-March of Dimes.

Eldridge, R.: The Torsion Dystonias: Literature Review: Genetic and Clinical Studies. In The Torsion Dystonias (Dystonia Musculorum Deformans). Editor, Roswell Eldridge, Neurology suppl. 20:11, Part 2, November 1970.

Eldridge, R. and Koerber, T.: The Torsion Dystonias: Some Genetic and Psychiatric Implications. The Psychiatric Forum, April, 1971.

Serial No. NDS(CF) - 67 E 1487
1. Collaborative & Field Research
2. Epidemiology Branch
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Genetic analysis of family data on Guam ALS and PD cases

Previous Serial Number: Same

Principal Investigators: Dwayne M. Reed, M.D.
Jacob A. Brody, M.D.
E. Michael Holden, M.D.
NINDS Research Center
Roswell Eldridge, M.D.

Other Investigators: Jose Torres
NINDS Research Center
Manuel T. Cruz
NINDS Research Center
Francisco Leon Guerrero
NINDS Research Center

Cooperating Units: NINDS Research Center, Tamuning, Guam
Center for Demographic and Population Genetics,
University of Texas at Houston (Dr. William J. Schull)

Man Years:

Total:	1
Professional:	1/4
Other:	3/4

Project Description:

Objectives: To utilize the accumulation of 20 years of experience for an indepth genetic analysis of pedigree information of Guam ALS and PD.

Methods employed: As earlier pedigree studies failed to disclose a simple autosomal mode of inheritance for ALS and PD, a number of other approaches are being continued and updated. One effort involves two registers based upon the first 100 deaths each for ALS and PD and an equal number of matched control deaths. All live-born offspring from these cases and controls have been registered and are being followed for the development of ALS or PD as well as death from all causes. A second effort involves the updating of a family registry started in 1958. This registry consists of 136 patients with either ALS or PD seen at the NINDS Research Center between 1958 and 1963, and an equal number of matched controls. All known first-degree

relatives and spouses of these cases and controls were registered by 1963 and all living relatives were examined at that time. Currently, the status of all relatives known to be living in 1963 is being updated for the development of ALS or PD and death from all causes. Another study involves the conjugal instances of ALS and PD. To date there have been 10 matings of ALS x ALS, 3 matings of PD x PD, and 14 matings of ALS x PD. The status of all offspring of these matings is currently being traced.

In cooperation with the Center for Demographic and Population Genetics, University of Texas at Houston, we have begun a computer genetic analysis for affected families. Since 1945 as each new case of ALS or PD has been seen at the NINDS Research Center, a family pedigree form has been completed. To date we have over 560 such pedigrees. Individual index cards have been maintained on each relative and this file is currently being prepared for computer analysis. In addition, a detailed tracing of the entire village of Umatac back to 1835 is being completed, utilizing church and vital statistic records.

Major findings: Genetic causation has been the most consistently involved explanation for the apparent familial concentration of ALS and PD among the Chamorros of Guam and other Mariana Islands. Autosomal models have difficulty accounting convincingly for the sex differences in incidence and mortality rates of ALS and PD. Sex-linked models are also unattractive for a number of reasons. For example, affected women do not always have affected fathers and affected fathers with normal spouses do not have only affected daughters as a sex-linked model requires.

For these reasons a number of new approaches have been undertaken to clarify the genetic issue. These steps include a careful search for the missing women which would account for the uneven sex distribution. This includes analysis of mortality records to determine whether women are dying of another cause which may be related to ALS or PD although phenotypically different. Secondly, an effort is being made to evaluate the fertility of individuals with one or both of these neurologic disorders. Complete ascertainment of the status of offspring of the first 100 cases of ALS and PD, the original family registry, and of the conjugal matings together with data from previous studies should allow a good comparison of the frequency of illness among the offspring of two affected, one affected, and no affected parental matings. Such information should be of considerable value in understanding the population dynamics of these diseases if indeed they are inherited.

Significance to biomedical research and the program of the Institute: Establishing a genetic basis for Guam ALS and PD would have far-reaching consequences. The elusive question of the cause of Guam ALS and PD would be answered. A new genetic disease would be added to the expanding catalog of inherited neurologic diseases.

Proposed course: Complete information is being obtained on pedigrees, fertility, and cause of death of relatives of cases and controls. When

the family registries and pedigrees updating has been completed analysis will be made of the genetic patterns of these diseases.

Honors and Awards: None

Publications: None

Serial No. NDS (CF) - 67 E 1488
1. Collaborative & Field Research
2. Epidemiology Branch
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Serological studies of common viruses in cases of multiple sclerosis (MS) and controls

Previous Serial Number: Same

Principal Investigators: Jacob A. Brody, M.D.
John L. Sever, M.D.
Infectious Diseases Branch, C&FR, NINDS
Anne H. Edgar

Other Investigator: None

Cooperating Units: Infectious Diseases Branch, C&FR, NINDS

Man Years:

Total:	1
Professional:	1/2
Other:	1/2

Project Description:

Objectives: To test the hypothesis that MS may be caused by an unusual response to a common virus infection. To search for possible distortions of segregation and association among MS patients, siblings and controls.

Methods employed: MS patients known to the Neurology Department, IUMC and to the Indiana Chapter of the National Multiple Sclerosis Society were contacted and asked to participate. In addition, for each case several controls with similar backgrounds and infectious disease experience were selected. Controls are classmate friends of the patient who grew up in the same community. Siblings of MS patients were also tested. A second sample of MS patients and siblings was taken from the Washington, D.C. area. Patients and controls answered standard questions regarding infectious disease, environment, course of illness and family history, and blood specimens were taken.

Serological analysis was conducted in the Infectious Diseases Branch, C&FR, NINDS using a battery of common virus antigens by hemagglutination and complement fixation methods. Differences between the patient, his sibling and his controls were analyzed.

A portion of frozen serum is being banked to test promising hypotheses in the future.

Major findings: In the Indiana series we found that MS patients have higher titers than matched controls for measles, mumps, influenza C, parainfluenza type 3, varicilla and herpes virus hominus. In no case, however, were titers of MS patients higher than their siblings of the same sex. In the Washington series, female siblings had titers as high as female patients to measles while male siblings titer were lower than those of the patients.

Significance to biomedical research and the program of the Institute: The observation that MS patients do have consistently higher titers against many viruses than controls supports an infectious or immune mechanism as being involved in the etiology of MS. The finding that higher titers also occur in siblings suggests that the phenomenon may be related to a common familial exposure or a familial immunologic defect.

Proposed course: We plan to extend our studies to the Shetland and Orkney Islands where MS occurs at a rate three times higher than elsewhere in the world and to use our current specimens for other studies as indicated.

Honors and Awards: None

Publications:

Henson, T.E., Brody, J.A. and Sever, J.L.: Elevated measles antibodies in patients with multiple sclerosis and in their siblings. Presented at the 97th Annual Meeting of the American Public Health Association, Philadelphia, Pa., November 1969.

Henson, T.E., Brody, J.A., Sever, J.L., Dyken, M.L. and Cannon, J.M.: Measles antibodies in patients with multiple sclerosis and their siblings and controls. JAMA, 211:1985, 1970.

Brody, J.A.: Virus antibody titers in multiple sclerosis patients, siblings and controls. (An abstract.) (Presented at the American Epidemiological Society Meeting in Seattle, Washington, April 1970.)

Brody, J.A., Sever, J.L. and Henson, T.E.: Virus antibodies in the serum of multiple sclerosis patients and matched controls. Neurology, 20:389, 1970. (Presented at the American Academy of Neurology, May 1970. Proceedings to be published.)

Brody, J.A., Sever, J.L. and Henson, T.E.: Virus antibodies in MS patients, siblings and controls. JAMA, 216:1441-1446, 1971.

Brody, J.A., Sever, J.L., Edgar, A.H. and McNew J.: Measles antibody titers of multiple sclerosis patients and their siblings. Neurology, 22:492-499, 1972.

Brody, J.A.: Epidemiologic and serologic data on multiple sclerosis and their possible significance. Presented to the Kroc Foundation Symposium on "Research in Multiple Sclerosis," in Santa Ynez, Calif., February 1972.

Brody, J.A.: Comments on the epidemiology of multiple sclerosis and a possible virus etiology. Lancet, II:173, 1972.

Brody, J.A.: Epidemiologic and serologic data on multiple sclerosis and their possible significance. In Multiple Sclerosis: Immunology, Virology, and Ultrastructure. UCLA Forum in Medical Sciences, #16. Eds. F. Wolfgram, G.W. Ellison, J.G. Stevens and J.M. Andrews. Chap. 6, pp. 127-141. Academic Press, New York, 1972.

Serial No. NDS (CF) - 67 E 1490
1. Collaborative & Field Research
2. Epidemiology Branch
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Development of an *in vitro* serum cytotoxicity assay for amyotrophic lateral sclerosis

Previous Serial Number: Same

Principal Investigators: Jacob A. Brody, M.D.
George Nemo, Ph.D.

Other Investigator: None

Cooperating Unit: None

Man Years:

Total:	1/8
Professional:	1/16
Other:	1/16

Project Description:

Objectives: To develop a simplified diagnostic test for amyotrophic lateral sclerosis (ALS).

Methods employed: The technique recently described by Dr. Frederick Wolfgram involves treating explant cultures of anterior horn cells of 3-day-old mice with diluted ALS serum. ALS serum appears to specifically destroy the neurons in culture. Toxicity is determined by visually examining strained preparations (Holmes and Bodian staining methods) using phase-contrast and regular light microscopy. We are trying to duplicate this work and to simplify the technique.

Major findings: None as yet.

Significance to biomedical research and the program of the Institute: It is hoped that the technique when applied as an epidemiologic tool will serve to elucidate the pathogenic event(s) leading to the disease.

Proposed course: We are attempting to set up the technique in our laboratory to further evaluate the specificity of the serum factor. We are particularly concerned with simplifying the system so that it may be used to test large groups of subjects with relative ease.

Honors and Awards: None

Publications: None

Serial No. NDS (CF) - 67 E 1496
1. Collaborative & Field Research
2. Epidemiology Branch
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Sequelae of CNS diseases in childhood and perinatal period

Previous Serial Number: Same

Principal Investigator: Milton Koch, M.D.

Other Investigator: Jacob A. Brody, M.D.

Cooperating Unit: University Hospital, University of West Virginia Medical Center, Morgantown, West Virginia

Man Years:

Total:	3/16
Professional:	1/16
Other:	1/8

Project Description:

Objectives: During this year we studied possible birth abnormalities of the CNS in relationship to a community-wide epidemic of diarrhea caused by a potentially teratogenic (parvovirus) virus in Morgantown, West Virginia which had occurred in May 1971.

Methods employed: All medical records in the University Hospital from 1967 until July 1972 coded for any complication of pregnancy, stillbirth, or congenital malformation were pulled and reviewed. Baseline numbers for normal deliveries were also obtained for that time period. The annual numbers of complications were recorded.

The numbers were compared in order to see whether there was a rise in any of these aforementioned complications related temporarily to the epidemic. This was considered possible because parvoviruses, which are small DNA viruses, are known to cause spontaneous abortion in cattle, and several other intrauterine complications, in addition to diarrheal illness in adult humans.

Major findings: After reviewing approximately 350 charts for the 5-year period, no increase in incidence of complications of pregnancy, stillbirth, or congenital malformations were noted for the 9-month period following May 1971.

Significance to biomedical research and the program of the Institute: From this brief study we can say that the parvovirus associated with this diarrheal epidemic did not produce major abnormalities which were grossly detectable at birth.

Proposed course: We will look for other situations where large groups of people were affected by viral illness to determine possible affects in utero and in early childhood.

Honors and Awards: None

Publications: None

Serial No. NDS (CF) - 68 E 1594
1. Collaborative & Field Research
2. Epidemiology Branch
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Phenothiazine-induced neurological effects: A study among twins

Previous Serial Number: Same

Principal Investigators: James A. Schnur, M.D.
Jacob A. Brody, M.D.

Other Investigators: None

Cooperating Units: National Institute of Child Health and Human Development, Children's Diagnostic and Study Branch
National Institute of Mental Health
Section on Twin & Sibling Studies, Adult Psychiatry Branch
National Academy of Science, National Research Council
Columbia University, New York State Psychiatric Institute
Spring Grove State Hospital, Catonsville, Maryland

Man Years:

Total:	3/16
Professional:	1/16
Other:	1/8

Project Description:

Objectives: The objective of this study is to assess whether the specific types of neurological side reaction induced by phenothiazine drugs are influenced by genetic factors.

Methods employed: A preliminary study among 46 female geriatric patients on long-term phenothiazine treatment revealed 33% had dyskinetic reactions and 13%, Parkinson-like reactions. The subjects for this study are twin pairs, concordant for the same psychiatric diagnosis, who have been on chronic phenothiazine therapy. Zygosity of the twin pairs is determined by history, appearance, and extensive blood typing. A single neurological examination was conducted on each pair to determine the patterns of neurological reactions. By comparing the patterns of reactions in monozygotic with those of fraternal twins, we can employ the usual methods of analysis to determine the relative importance of genetic factors in the manifestation of extrapyramidal signs resulting from phenothiazine induction.

Major findings: Six pairs of twin patients, four monozygotic and two dizygotic, on long time-high dosage phenothiazine treatment have been examined. The results indicated that the specific type of neurological side effect is not primarily determined by genetic factors, since identical twins may develop distinctly different patterns of reaction.

Significance to biomedical research and the program of the Institute: This work may help elucidate the presence or absence of a genetic contribution to the occurrence of the important neurological side reactions to phenothiazine drugs.

Proposed course: We propose to add other twin pairs to the series, and analyze the data.

Honors and Awards: None

Publications: None

Serial No. NDS (CF) - 68 E 1597
1. Collaborative & Field Research
2. Epidemiology Branch
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Neurologic diseases in the Trust Territories and other Pacific areas

Previous Serial Number: Same

Principal Investigators: Jacob A. Brody, M.D.
E. Michael Holden, M.D.
NINDS Research Center
Dwayne Reed, M.D.

Other Investigators: Manuel Cruz
NINDS Research Center
Jose Torres
NINDS Research Center
Francisco Leon Guerrero
NINDS Research Center

Cooperating Unit: None

Man Years:

Total:	1 1/4
Professional:	1/4
Other:	1

Project Description:

Objectives: To investigate neurologic illness occurring in the Trust Territories and the Pacific area. During the past few years, this involved the study of leprosy patients in New Caledonia, and congenital blindness among the people of Pingelap. Currently we are looking for ALS and PD among non-Chamorro groups.

Methods employed: Through contact with the Department of Health, U.S. Trust Territory and other agencies involved in health in this area, we continue to search for unusual patterns of neurologic disease.

Major findings: We are currently following several Carolinian patients on Saipan suspected of having ALS. In addition, we have hospital discharge diagnoses of ALS for two patients in the Truk district and one from Palau district. Reports from a health survey in the 1950's were uncovered and 10 patients in a village of 245 in Palau were reported as having parkinsonism. We plan to follow up these reports.

We also continue to follow the congenital eye disease problem in Pingelapese.

Significance to biomedical research and the program of the Institute: By discovering population isolates with unusual patterns of neurologic disease, we have unusual opportunities to study the mechanisms of cause and control.

Proposed course: We will continue to follow this population and other populations as they become known which are of potential medical interest.

Honors and Awards: None

Publications: Brody, J.A., Hussels, I., Brink, E. and Torres, J.M.: A preliminary report on hereditary blindness among the Pingelapese people of the Eastern Caroline Islands. Lancet, I:1253-1257, 1970.

Stanhope, J.M.: Pingelapese migrant communities of Ponape, Eastern Caroline Islands. Human Biology in Oceania. In press.

Serial No. NDS (CF) - 68 E 1598
1. Collaborative & Field Research
2. Epidemiology Branch
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Epilepsy on Guam

Previous Serial Number: Same

Principal Investigator: Jacob A. Brody, M.D.

Other Investigators: Manuel Cruz
NINDS Research Center
Jose Torres
NINDS Research Center
Francisco Leon Guerrero
NINDS Research Center
Kwang-Ming Chen, M.D.
Guam Memorial Hospital

Cooperating Unit: NINDS Research Center, Tamuning, Guam

Man Years:

Total:	13/16
Professional:	1/16
Other:	3/4

Project Description:

Objectives: To determine the incidence and prevalence of epilepsy on Guam. To investigate methods for field studies of epilepsy. To determine if previous reports of unusually high incidence of convulsive disorders on Guam are accurate.

Methods employed: We are testing four basic approaches: (1) We are following-up a 1962 survey of convulsive disorders in Umatac and Merizo to determine the outcome of those children known to have had febrile convulsions 6 to 8 years ago; (2) to determine the true incidence and prevalence in a sample population we are doing a house to house survey in the villages of Talofofo, Merizo, and Yona; (3) as referral neurologists on Guam we are updating all previous referrals of convulsive disorders to us and establishing a registry. To add to this registry we are contacting all medical and paramedical personnel on Guam to discover new cases. This registry will be permanent and permit us to conduct studies in the Guam population; (4) to further elaborate on methods for acquiring information we are following-up all births on Guam in 1958 and 1963 throughout the entire island to determine the rates of convulsive disorders in these preselected populations.

Major findings: Epilepsy rates were slightly higher on Guam than in most areas where data exists. We found that clinical and hospital sources must be supplemented by field surveys to approach a "true" rate. While symptomatic epilepsy was fairly well reported through medical channels idiopathic grand mal epilepsy was severely underreported. Febrile convulsions were inadequately reported through medical channels. While field surveys were more complete our best method for detection was by follow-up of birth cohorts for given years. Rates were consistently higher in some areas but no genetic or environmental factors emerged to explain this.

Significance to biomedical research and the program of the Institute: Further studies of epilepsy in a well-defined and accessible population will add to the understanding of this disease and contribute new information concerning epilepsy in a tropical environment. It will also yield important information on different survey techniques and their relative accuracy.

Proposed course: The registry will be maintained on Guam for future studies.

Honors and Awards: None

Publications: Stanhope, J.M., Brody, J.A. and Brink, E.: Convulsions among the Chamorro people of Guam, Mariana Islands. I. Seizure Disorders. Amer. J. Epid. 95:292-298, 1972.

Stanhope, J.M., Brody, J.A., Brink, E. and Morris, C.E.: Convulsions among the Chamorros people of Guam, Mariana Islands. II. Febrile Convulsions. Amer. J. Epid. 95: 299-304, 1972.

Serial No. NDS (CF) - 68 E 1605
1. Collaborative & Field Research
2. Epidemiology Branch
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Phenothiazine-induced parkinsonism in white and Negro patients with nonorganic psychoses

Previous Serial Number: Same

Principal Investigators: James A. Schnur, M.D.
NINDS Research Center
R. Michael Scott, M.D.

Other Investigators: Jacob A. Brody, M.D.
Joyce Cannon, R.N.

Cooperating Units: Spring Grove State Hospital, Catonsville, Maryland
Crownsville State Hospital, Crownsville, Maryland

Man Years:

Total:	3/16
Professional:	1/16
Other:	1/8

Project Description:

Objectives: To determine whether increased skin pigmentation is associated with decreased prevalence of phenothiazine-induced Parkinson-like syndromes.

Methods employed: This project was originally designed to investigate influence of skin pigmentation on the prevalence of naturally occurring Parkinson's disease by use of a questionnaire which was to be mailed to a sample of American physicians. A pilot study, however, showed this approach to be impractical, and therefore, it was abandoned. In view of the possible relationship between drug-induced and naturally occurring parkinsonism, we then decide to study comparable white and Negro populations who were on treatment with phenothiazines. Thus far, we have examined approximately 75 patients in each group, samples sufficient in number for a comparative analysis.

Major findings: Our initial results suggest that there is no difference in the prevalence of phenothiazine-induced parkinsonism between the two populations surveyed. We are still working out methods to evaluate the effects of duration and type of medication.

Significance to biomedical research and the program of the Institute: To further define the relationship between melanogenesis in the skin and in the pigmented nuclei of the basal ganglia.

Proposed course: Analysis of data is now in progress.

Honors and Awards: None

Publications: Brody, J.A.: Genetic considerations in Parkinson's disease. Presented at the Laurentian L-Dopa Conference, Montreal, November 1969. In: L-Dopa and Parkinsonism, Edited by Andre Barbeau and Fletcher McDowell, F.A. Davis Co., Phila., 1970, pp. 27-30.

Serial No. NDS (CF) - 69 E 1774
1. Collaborative & Field Research
2. Epidemiology Branch
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Neurologic signs and symptoms associated with malabsorption

Previous Serial Number: Same

Principal Investigators: Milton J. Koch, M.D.
Jacob A. Brody, M.D.
Anne H. Edgar

Other Investigator: Paul M. Hoffman, M.D.

Cooperating Units: Central Veterans Administration Authority, Washington, D.C.
Veterans Administration Hospital, Atlanta, Georgia
Veterans Administration Hospital, Long Beach, California
Veterans Administration Hospital, Durham, North Carolina
Veterans Administration Hospital, Los Angeles, California

Man Years:

Total:	5/16
Professional:	1/16
Other:	1/4

Project Description:

Objectives: To determine if both clinical and subclinical malabsorption is associated with a high incidence of neurologic signs and symptoms.

Methods employed: The Cooperative Veterans Administration Retrospective Study of surgery for peptic ulcer served as the source for patients in this study. A review of abstracts from this study showed that a given hospital was more likely to have done surgical procedure for the majority of its cases then another. For this reason, patients with 3 different surgical procedures performed at 4 hospitals were chosen. A fourth group of patients who had simple closures of perforated ulcers were selected from all 4 hospitals. Patients who had a subtotal gastrectomy with a Billroth II reanastomosis were selected from the Durham Veterans Hospital, patients who had a hemi-gastrectomy and vagotomy were selected from the Atlanta Veterans Hospital, and patients who had a vagotomy and pyloroplasty were selected from the Long Beach and Wadsworth Veterans Hospital. In order to insure that this population would be in the most susceptible age group, patients between the ages of 30 and 80 who had had surgery between 1952 and 1957 were selected from abstracts and hospital records where available. Patients were not

included in the study if there was a history of carcinoma, severe diabetes with neurologic complications, any systemic disease with known neurologic complications, or a history of neurologic disease prior to their ulcer surgery. Patients were also excluded from the study if a revision of their original ulcer surgery or subsequent surgery for a recurrent ulcer had been performed. No attempt was made in this study to analyze the causes of death or those who died since surgery. A mortality study of the 2,800 original cases which includes this sample has also been undertaken. All patients included in the study had letters sent to their last known address asking them to report on one of several days to the outpatient clinic of the hospital where their surgery was performed. A complete history with special attention on the neurologic and gastrointestinal systems as well as a complete neurologic examination was performed on all patients and a random sample of all patients as well as all patients with abnormal findings were examined by a staff neurologist.

Major findings: No cases of motor neuron disease have been found in the examined group. A large number of cases of peripheral neuropathy were identified in the vagotomy and hemi-gastrectomy group. Only two cases were seen in the vagotomy and pyloroplasty group. Thorough evaluation of all patients with unexplained neurologic disorders in the Atlanta group showed that malabsorption, poor nutrition, chronic alcohol intake, and weight loss was all more prevalent in this group than in those without neurologic findings. In reviewing 763 death certificates obtained by follow-up of 2500 cases of the original study group we encountered 2 patients with multiple sclerosis, 2 with Parkinson's disease and 1 with amyotrophic lateral sclerosis. This group was updated again this year and we are awaiting final outcome.

Significance to biomedical research and the program of the Institute: There have been many clinical reports of cases of malabsorption who have shown signs and symptoms of nervous system disease. There have also been scattered reports in literature of patients who are known to have motor neuron disease, who had a history of having had a Billroth II type of gastric surgery performed many years prior to the onset of their disease. There has never been, however, a matched, controlled population study of the association of these two abnormalities. If this association is valid then further study into the mechanism of absorption of essential nutrients and their incorporation into nervous tissue may be a meaningful approach to the study of chronic neurologic disease.

Proposed course: This project will continue with emphasis on causes of death within this population. At the present time the status of an additional 392 cases of the original study group is being sought in order to extend the information on the causes of mortality in this population.

Honors and Awards: None

Publications: None

Serial No. NDS (CF) - 69 E 1777
1. Collaborative & Field Research
2. Epidemiology Branch
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: ALS among non-Chamorros after residence on Guam

Previous Serial Number: Same

Principal Investigator: Jacob A. Brody, M.D.

Other Investigator: Estelle Kornhauser, R.N.
Dwayne Reed, M.D.
Anne H. Edgar

Cooperating Unit: Bureau of Data Processing and Accounts, Social Security Administration, DHEW

Man Years:

Total:	9/16
Professional:	1/16
Others:	1/2

Project Description:

Objectives: This project was developed to determine if prolonged exposure (over one year) to the environment of Guam increases the likelihood of the development of ALS among statesiders.

Methods employed: Through various workers in the Department of Defense we were put in contact with several construction companies which had maintained large staffs of statesiders on the island of Guam after World War II. These companies were asked to supply us with the names, birthdates, and social security numbers of all personnel employed on Guam and from these lists we selected only those workers who had spent more than one year on Guam. This was a group of approximately 12,000 individuals. The Social Security Administration searched its records to determine which of these workers had died and where they had died. We are contacting the individual states to obtain the death certificates of the deceased workers and determine the cause of their death.

Major findings: Of the approximately 12,000 cards submitted to the Social Security Administration, 450 were found to contain incorrect information making follow-up impossible. 7,730 were considered as representing individuals still alive, and the remainder (4,000) were identified as to place of death. The names were submitted to the individual states and death certificates requested. About 90% of the requested

certificates have been returned to date. Preliminary analysis indicates that there was no excess of ALS in this group.

Significance to biomedical research and the program of the Institute: Although results on this study are far from complete, the initial trend is that the rate of ALS among statesiders who have spent considerable time on Guam remains the same as that for statesiders who have never spent time in that environment. If this trend is borne out by subsequent findings, it would suggest that the environmental factors on Guam are less likely to be responsible for high rate of ALS among its native population.

Proposed course: All data are coded and we are awaiting computer runs for final analysis.

Honors and Awards: None

Publications: None

Serial No. NDS (CF) - 69 E 1779
1. Collaborative & Field Research
2. Epidemiology Branch
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: The epidemiology of motor neuron disease in the United States

Previous Serial Number: Same

Principal Investigators: Jacob A. Brody, M.D.
Anne H. Edgar

Other Investigator: None

Cooperating Unit: None

Man Years:

Total:	9/16
Professional:	1/16
Other:	1/2

Project Description:

Objectives: To determine death rates due to amyotrophic lateral sclerosis among native-born and migrant residents of California and Washington.

Methods employed: Certificates were collected for all deaths attributed to ALS as primary or underlying cause, occurring in California residents from 1954 to 1964, excluding 1956, and in Washington residents from 1955 to 1964. Death certificates were analyzed for age at death, sex, race, place of residence and place of birth. The material presented was limited to American-born whites. Death rates for native-born Californians and Washingtonians and for migrants to these states from the nine geographical divisions of the United States were calculated from the average number of deaths per year by region of birth and the corresponding population at risk. Because the age and sex composition of the migrant and native populations differed, the death rates were adjusted for age and sex using the population of the United States in 1960 as a standard.

Major findings: During the ten-year period under study, the average age-sex-adjusted ALS death rates were similar for the native-born populations of California and Washington (0.62 and 0.57 per 100,000 respectively). Rates among American-born migrants from southern and northern areas to California and Washington ranged from .74 to 1.03 for males and .39 to .60 for females.

There was no correlation of ALS death rates with state or area of birth as we had demonstrated in a parallel study of MS in the same populations. The results provide no suggestion of possible exogenous factors which may be related to the etiology of amyotrophic lateral sclerosis.

Significance to biomedical research and the program of the Institute: Numerous epidemiologic studies of mortality from amyotrophic lateral sclerosis have attempted to uncover patterns of disease distribution which might provide a suggestion as to the etiology of this disease. The great proportion of this research has been based on national and international mortality statistics and, therefore, interpretation of the results must take into account geographic variations in medical care, diagnostic facilities, coding of deaths and death certification. The design of this study allows for a comparison of rates among people born in various areas of the country but who were subject to similar standards of medical care and certification at the time of death. The results suggest that future research into the causes of amyotrophic lateral sclerosis might emphasize other than those related to geographic location.

Proposed course: Further studies of epidemiologic patterns of ALS are contemplated.

Honors and Awards: None

Publications: Edgar, A.H., Brody, J.A. and Detels, R.: Amyotrophic lateral sclerosis mortality among native-born and migrant residents of California and Washington. Neurology, 23:48-51, 1973.

Serial No. NDS (CF) - 69 E 1780
1. Collaborative & Field Research
2. Epidemiology Branch
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1971 through June 30, 1972

Project Title: Familial bilateral acoustic neuroma

Previous Serial Number: Same

Principal Investigator: Roswell Eldridge, M.D.

Other Investigators: Dean F. Young, M.D
New York, New York
Henry Hood, M.D.
Danville, Pennsylvania
W. J. Gardner, M.D.
Cleveland, Ohio
George T. Nager, M.D.
John Hopkins Hospital
Frank H. DeLand, M.D.
John Hopkins Hospital

Cooperating Units: Department of Otolaryngology,
Department of Radiology,
Department of Neurosurgery,
Geisinger Medical Center
Danville, Pennsylvania
John Hopkins Hospital
Baltimore, Maryland

Man Years:

Total:	3/8
Professional:	1/8
Other:	1/4

Project Decription:

Objectives: To perform genetic, clinical and physiologic studies of a large family with hereditary bilateral acoustic neuroma.

To clarify the relationship of this trait to other disorders with acoustic neuromas such as neurofibromatosis.

Methods employed: Field studies were conducted in evaluating family members. On those seen personally, physical examinations were performed, stressing neurological and skin examinations. In addition, audiometric examinations including air and bone conduction and caloric examinations

were conducted in the field.

Genealogic information was obtained from family members, family records, D.A.R. records, state and military records, census recordings. Medical history was obtained from family members, hospital and physician records, occasionally from school records or military records.

On select patients extensive outpatient studies have included audiometric and vestibular testing, complete ENT and neurologic examinations, skull x-rays and brain scans using radioactive techniques. These were undertaken in cooperating with the Department of Neurology, Radiology, and Otolaryngology of John Hopkins Hospital.

Major findings: The study of this, the largest kindred with hereditary neoplasm yet reported, has generated information which has been particularly useful to neurosurgeons and otolaryngologists since it suggests a relatively benign prognosis but high prognosis and genetic risk to those individuals who develop such a disease at a young age.

Significance to biomedical research and the program of the Institute: The mode of inheritance to this trait has been confirmed and the place of this syndrome among those associated with neural sheath proliferation is clearer. Of primary importance is the establishment of appropriate diagnostic technique for early cases and treatment of these cases.

Proposed course: This Project is Terminated.

Honors and Awards: None

Publications:

Young, D.F., McNew, J. and Eldridge, R.: Hereditary Acoustic Neuroma-Clinical and Genetic Aspects. In Transactions of the American Neurological Association. 94, 1969.

Young, D.F., Eldridge, R., Nager, G.T., DeLand, F.H., and McNew, J.: Hereditary bilateral acoustic neuroma (central neurofibromatosis). Proceedings of the Second Conference on The Clinical Delineating of Birth Defects, May 1969. The National Foundation-March of Dimes.

Young, D., Eldridge, R., and Gardner, W.J.: Bilateral Acoustic Neuroma in a Large Kindred. JAMA. 214, October 1970

Serial No. NDS (CF) - 69 E 1781
1. Collaborative Field Research
2. Epidemiology Branch
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1971 through June 30, 1972

Project Title: Twin Studies in Parkinson's Disease

Previous Serial Number: Same

Principal Investigators: Roswell Eldridge, M.D.
Zdenek Hrubeck, M.D.
National Academy of Sciences

Other Investigator: Kathy O'Meara

Cooperating Unit: The National Academy of Sciences - National Research Council Twins Registry, Washington, D. C.

U. S. Veterans Administration
Washington, D. C.

Man Years:

Total:	1/8
Professional:	1/16
Other:	1/16

Project Description:

Objectives: Parkinsonism is not a single entity but rather a symptom complex which may be idiopathic, or may be due to cerebral arteriosclerosis or may follow encephalitis. Genetic factors have been considered important by a number of authors but it is not yet established how important such factors are in any one form of Parkinsonism or if there is a form which has a simple genetic basis independent of acquired neurologic disease. The aim of the present proposal would be to use the twin method to evaluate genetic factors in various forms of Parkinsonism and/or to distinguish genetically determined Parkinsonism from other forms.

Methods employed: Survey of the VA twin registry in 1968 has revealed eight cases of Parkinsonism in one of a Veteran twin pair. Each of the pair were apparently discordant for Parkinsonism. Two twin pairs were monozygotic, five were dizygotic and in one the zygosity was unknown. We would contact each of these individuals and his co-twin, and others with the diagnosis who may be ascertained through updating of the registry, and arrange for an appointment with the individual in his home at a time when available relatives could also be present. During the family interview

a pertinent medical history would be obtained and physical examination would be performed. Permission for review of hospital records would be secured and arrangements might be made for additional neurologic studies. To define zygosity, photographs would be taken of the twins, blood would be drawn for genotyping and dermatoglyphics might be recorded. (Personal examination of both twins and available relatives is important in order that mild cases not be missed).

Major findings: Interest continues in this project although there has been no additional ascertainment of cases since the last report. It has not yet been possible to contact patients through the National Science Foundation registry but we expect twin pairs will be available to us in the future.

Significance to biomedical research and the program of the Institute: In a chronic condition with late onset it is often difficult to determine the role of genetic factors in causation. The twin method provides a relatively simple method involving small numbers to answer this question.

The nosology of Parkinson's disease is especially important now that the drug L-Dopa has been shown to help some with Parkinsonism. Is this drug most helpful in a specific form of the disease? Is it effective in hereditary Parkinsonism?

Proposed course: This project is terminated.

Honors and Awards: None

Publications: None

Serial No. NDS (CF) - 69 E 1782
1. Collaborative & Field Research
2. Epidemiology Branch
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1971 through June 30, 1972

Project Title: Twin Studies in Torticollis

Previous Serial Number: Same .

Principal Investigators: Roswell Eldridge, M.D.
Zdenek Hrubec, M.D.
National Academy of Sciences

Other Investigators: Kathy O'Meara

Cooperating Unit: The National Academy of Sciences - National Research Council Twins Registry, Washington, D.C.

U.S. Veterans Administration
Washington, D.C.

Man Years:

Total:	1/8
Professional:	1/16
Other:	1/16

Project Description:

Objectives: Torticollis may be broadly divided into infantile and post-infantile forms. The former may be congenital or appear several weeks after birth but in either the cause appears due to events preceding birth. Post-infantile torticollis consists of a heterogeneous group of disorders which have been ascribed to a number of causes including trauma or inflammation of the cervical spine, myositis or nuchal musculature, functional illness, and disease of the peripheral or central nervous system. In addition torticollis on a hereditary basic, either as an isolated symptom or in association with other movement disorders such as torsion dystonia, has been the subject of numerous reports. The aim of the proposed study is to weigh the genetic factors in the post-infantile forms of torticollis and, if possible, to distinguish between discrete hereditary types.

Methods employed: In 1968 a review of the Veterans Administration twin registry disclosed 11 individuals with a diagnosis of torticollis. Each of the 11 pair were said to be discordant. Five were monozygotic, two were dizygotic and in the four the zygosity could not be established. We would contact each of these individuals and his co-twin, arrange for an appointment with the individual in his home at a time when available relatives

could also be present. (We would hope also to ascertain new cases by assisting in the up-dating of the twin registry). During the family interview the pertinent medical history would be obtained for all relatives and physical examination performed on those present. Permission for review of hospital records would be secured and arrangements might be made for additional neurologic studies. To establish zygosity, photographs would be taken of the twins, blood drawn for genotyping, and dermatoglyphics might be recorded.

Major findings: Interest continues in this project although there has been made no additional ascertainment of cases since the last report. It has not yet been possible to contact patients through the National Science Foundation registry but we expect twin pairs will be available to us in the future.

Significance to biomedical research and the program of the Institute: Torticollis may be hereditary or acquired but under each of these headings there appear to be a number of discrete entities. The twin method presents a relatively simple means to distinquish genetically determined forms. Concentration on torticollis which is simply inherited is worthwhile since such disorders should have a discrete biochemical basis which might be revealed by study with the neurochemical techniques now available.

Proposed course: This Project is Terminated.

Honors and Awards: None

Publications: None

Serial No. NDS (CF) - 70 E 1832
1. Collaborative & Field Research
2. Epidemiology Branch
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Serologic responses of multiple sclerosis patients and controls to a virus isolated from a multiple sclerosis case

Previous Serial Number: Same

Principal Investigators: George Nemo, Ph.D.
Jacob A. Brody, M.D.

Other Investigator: Hilary Koprowski, M.D.
Wistar Institute

Cooperating Unit: The Wistar Institute, Philadelphia, Pennsylvania

Man Years:

Total:	1/2
Professional:	1/4
Other:	1/4

Project Description:

Objectives: To measure antibody levels in serum from MS patients and controls against MS virus 6/94 which was recovered from the brain of an MS patient to explore the relationship between the agent and MS.

Methods employed: A total of 48 patients, 24 females and 24 males, and an equal number of controls were included in the study. Each patient was matched with a control who was a lifelong friend of similar age and sex, who was born in the same community and who attended the same schools.

Serum antibody determinations using MS virus 6/94 and a classical parainfluenza Type I isolated from a patient with acute respiratory disease were carried out employing standard hemagglutination-inhibition (HI) techniques.

Major findings: We were unable to demonstrate higher HI antibody titers among MS patients and controls for the 6/94 MS isolate. Similarly, no differences were found in antibody to standard human parainfluenza I. The etiologic relationship of the virus isolate to MS could not be determined with this procedure.

Significance to biomedical research and the program of the Institute: It has long been hypothesized by numerous workers that MS may be due to a viral etiology. The recent isolations of an infectious agent related to

group I parainfluenza virus from co-culture of MS brain cells and indicator cells seemed to strengthen the hypothesis. It is the purpose of this study to determine the relationship of parainfluenza I virus, if any, to MS.

Proposed course: Alternative serological procedures are being employed using the same study group.

Honors and Awards: None

Publications: None

Serial No. NDS (CF) - 70 E 1833
1. Collaborative & Field Research
2. Epidemiology Branch
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Immune mechanisms in chronic and congenital viral infections

Previous Serial Number: Same

Principle Investigator: Lon R. White, M.D., DB, NICHD

Other Investigators: George Nemo, Ph.D.,
Jacob A. Brody, M.D.
Harrie Anne Sutton, biologist, DB, NICHD
Samuel Baron, M.D., LVD, NIAID

Cooperating Unit: Developmental Biology Branch, NICHD (HD DB - 5)

Man Years:

Total:	1/8
Professional:	1/16
Other:	1/16

Project Description:

Objectives: 1. To establish models of chronic, congenital, and/or latent viral infection in animals and in tissue culture. 2. In infected animals, to define the development of immunity to the infecting and to heterologous antigens. 3. In tissue culture, to define the effect of immunologic factors (antibodies, immune lymphocytes, interferon) on the evolution of acute and chronic infections.

Methods employed: Neonatal and pregnant mice are injected with either a reovirus type 1 or the minute virus of mice (MVM). The persistence of virus in animals is determined by isolation and fluorescent and immunofluorescent techniques. Serum levels of hemagglutination inhibition antibodies to both agents are followed. Lymphocyte transformation and cytotoxic activity is studied in spleen cell cultures in the presence of phytohemagglutinin on specific antigens. Interferon production is studied by assay of media from tissue culture preparations of cells infected with MVM, and by the ability of MVM infected cells to support the growth of other viruses. Sensitivity of MVM to interferon is tested by determining the yield of virus following exposure of cells in culture to known amounts of interferon.

Major findings: MVM does not appear to induce interferon elaboration in vitro. Pretreatment of L cells with interferon produces a mild to moderate diminution in virus yield.

Significance to biomedical research and the program of the Institute: Infection of the human embryo or fetus with rubella or cytomegalovirus is associated not only with developmental abnormalities and mental retardation, but also with infection persisting for months or years after, despite the presence of neutralizing antibody in the serum. There are several examples of related phenomena in experimental animals infected during prenatal or neonatal life. Previous studies have suggested that an impairment of the immune function of lymphocytes may be associated with such persistent infection. These phenomena provide insight into the question of how viral infection is normally terminated, and represent a challenge to older ideas on the role of specific cellular immunity in the natural history of virus infections. The investigation described represents a direct experimental approach to elucidating the cause and course of chronic infection following initial exposure to the agent during immunologic immaturity. The results of this investigation will be of immediate relevance to our understanding of other types of illness known or suspected to be associated with chronic viral infection. In addition, they may suggest new approaches to research in the role of congenital viral infection as a cause of diseases of unknown etiology such as prematurity, idiopathic growth failure, malignancy, mental retardation, developmental malformation, and autoimmune diseases.

Proposed course: To be continued.

Honors and Awards: None

Publications: None

Serial No. NDS (CF) - 70 E 1835
1. Collaborative & Field Research
2. Epidemiology Branch
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Epidemiologic and immunologic study of families of subacute sclerosing panencephalitis patients and families of matched controls

Previous Serial Number: Same

Principal Investigators: Roger Detels, M.D.
Jacob A. Brody, M.D.
Anne H. Edgar

Other Investigators: John Sever, M.D.
Head, Infectious Diseases Branch, C&FR, NINDS
J. Anthony Morris, M.D.
Director, Slow, Latent & Temperate Viruses Branch, Bureau of Biologics, FDA

Cooperating Units: Infectious Diseases Branch, C&FR, NINDS
Slow, Latent & Temperate Viruses Branch, Bureau of Biologics, FDA

Man Years:

Total:	9/16
Professional:	1/16
Other:	1/2

Project Description:

Objectives: To determine, through the use of a matched control study, factors related to the etiology of subacute sclerosing panencephalitis.

Methods employed: Cases of SSPE were selected from the areas around St. Louis, Missouri, North Carolina, and California in order to evaluate the occurrence in distinctly different environments. 42 families with matched controls and an additional 8 families for whom a suitable control could not be obtained have been interviewed. Controls were selected on the basis of close friendship and similar background to the proband. A detailed history of events possibly related to the etiology of SSPE including episodes of measles, unusual illnesses, exposure to sick animals, etc. was asked of probands, controls and their families. A 20cc sample of blood was drawn from each individual in the proband and control families.

Major findings: Analysis of the 42 cases indicates that the median age of measles among probands was 18 1/2 months, 2 years younger than the median age of measles among controls. One-quarter of the probands, but none of the controls, had measles under one year of age. One-quarter of the probands had no history of measles. Other significant associations were chickenpox occurring just prior to measles and a history of exposure to fowl and to dogs with possible distemper. No inner city patients were found except for 3 in New York who were brought up in Puerto Rico and 2 in Chicago who had extensive contact with rural environments. Measles titers were elevated only among patients and not their siblings. Measles titers in mothers of patients with early measles did not differ from control mothers.

Significance to biomedical research and the program of the Institute: We have shown that the measles infection in cases was unusual and occurred frequently during a time when passive immunity was present. Further SSPE occurs in an unusual epidemiologic pattern with rates among males far exceeding those of females and the absence of true inner city cases. Thus we have postulated that SSPE is caused by an unusual measles infection and a subsequent triggering event which is probably a zoonotic virus.

Proposed course: Intensive search for the possible "zoonotic trigger" will be carried out with appropriate groups when the situation becomes available. We are also collaborating with the Bureau of Biologics, FDA in developing a serologic test of canine distemper virus.

Honors and Awards: None

Publications:

Brody, J.A. and Detels, R.: Subacute sclerosing panencephalitis: A zoonosis following aberrant measles. Lancet, II:500-501, 1970.

Brody, J.A., Detels, R. and McNew, J.: Evidence that subacute sclerosing panencephalitis is caused by an aberrant measles infection followed by a zoonosis. Presented at the Annual Meeting of the American Academy of Neurology, April 1971. Neurology, April 1971. Neurology, 21:439, 1971.

Brody, J.A., Detels, R. and Sever, J.L.: Mealses antibody titers in sibships of subacute sclerosing panencephalitis patients and controls. Lancet, I:177-178, 1972.

Detels, R., Brody, J.A., McNew, J. and Edgar, A.H.: An epidemiologic study of subacute sclerosing panencephalitis suggesting an induced aberrant immunologic mechanism. In press.

Serial No. NDS (CF) - 70 E 1836
1. Collaborative & Field Research
2. Epidemiology Branch
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Epidemiologic factors in Creutzfeldt-Jakob disease

Previous Serial Number: Same

Principal Investigators: Jacob A. Brody, M.D.
A. Roger Bobowick, M.D.
Raymond P. Roos, M.D.
IR, NINDS

Other Investigator: Marjorie Matthews, R.N.

Cooperating Unit: Laboratory of Slow, Latent and Temperate Virus Infections, IR, NINDS

Man Years:

Total:	3/4
Professional:	1/4
Other:	1/2

Project Description:

Objectives: Creutzfeldt-Jakob disease is one of the spongiform encephalopathies which has been transmitted to chimpanzees. There are 3 forms of Creutzfeldt-Jakob disease which are distinct clinically. Nothing is known of the epidemiology of this syndrome. We wish to determine if there are common factors among patients who develop Creutzfeldt-Jakob disease and if there are specific factors related to distinct forms of this syndrome.

Methods employed: Letters were sent to neuropathologists soliciting pathologically proven cases and cases were used from the series at the Laboratory of Slow, Latent and Temperate Virus Infections, IR, NINDS. With the cooperation of the referring physicians the families of the patients are contacted and asked to participate in the epidemiologic interview. At the time of the interview session a detailed questionnaire was completed on the relative who is the interviewee, on the patient, and on an age and sex matched friend of the patient who serves as a control. Blood samples are also taken for future serologic studies.

Major findings: From the series of 69 patients, we have interviewed 38 families. The methods outlined above have proved workable. We found that patients with the classical or slowly progressive form of the disease

associated with amyotrophy are on an average 10 years younger than other Creutzfeldt-Jakob disease patients. This type of disease has not yet transmitted to chimpanzees. Severe upper respiratory illness merged with the onset of Creutzfeldt-Jakob disease in 3 patients with the ataxic form of Creutzfeldt-Jakob disease. No specific factors, exposures or prior history of disease distinguished the patients from the controls. It was of interest, however, that one-third of the patients and one-third of the controls ate animal brains. The patients tended to have eaten animal brains more frequently and more recently than controls. Also 10 of the 13 patients and only 3 of the controls stated specifically that they ate hog's brains.

Significance to biomedical research and the program of the Institute: This neurologic disease is caused by a virus or a combination of viruses. Transmissible diseases must be studied epidemiologically if predisposing factors are to be elicited. The documentation of a predisposing factor to a virus-induced degenerative disease would be a major contribution to the understanding of the disease, its treatment and prevention. While there was no overall difference in brain consumption between patients and controls, the information, particularly about hog brains, may be a lead since all the other slow virus transmissible spongiform encephalopathies are transmitted in nature through exposure to brain material.

Proposed course: We are now extending our study by using a modified questionnaire in additional patients and controls to get more information on consumption of animal brains and respiratory illness. We are also trying to secure information on animal brain consumption in the United States and Great Britain.

Honors and Awards: None

Publications: Bobowick, A.R., Matthews, M.A., Brody, J.A., Roos, R. and Gajdusek, D.C.: Creutzfeldt-Jakob Disease: A case-control study. In press.

Serial No. NDS (CF) - 70 E 1837
1. Collaborative & Field Research
2. Epidemiology Branch
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Multiple sclerosis distribution and patterns

Previous Serial Number: Same

Principal Investigators: Jacob A. Brody, M.D.
Dwayne Reed, M.D.
Milton Koch, M.D.

Other Investigators: Marjorie Matthews, R.N.

Cooperating Units: None

Man Years:

Total:	1/2
Professional:	1/4
Other:	1/4

Project Description:

Objectives: To determine the role of geographic latitude in the risk of multiple sclerosis among migrants. Conduct epidemiologic investigations of unusual clusters of cases of multiple sclerosis. To study possible risk factors before age 20 which distinguish cases from controls.

Methods employed: This project utilizes collection and analysis of mortality records from high and low risk areas, and morbidity studies of affected persons and controls in stable communities, and routine epidemiologic investigations of clusters of multiple sclerosis cases.

Major findings: Earlier completed aspects of this study have indicated that multiple sclerosis mortality rates among migrants are a function both of the geographic latitude of origin and destination. Migrants to California and Washington State from areas reported to have low rates of multiple sclerosis had themselves low mortality rates, suggesting that they had acquired protection prior to migration. Migrants to Washington State from areas reported to have high rates had, themselves, higher rates than similar groups of migrants to California, suggesting that the "protective" factor of southern latitudes was operative after migration.

We now know of five apparent foci of multiple sclerosis, one in Comanche County, Kansas, a second in Mossyrock, Washington, a third in Superior,

Wisconsin, a fourth in Danbury, Connecticut, and a fifth in suburban Boston. We have partially worked up the first 2 and intend to continue with descriptive studies of the other areas, in hopes of finding common environmental factors. In Comanche County we know of 7 cases in a population of 3,000 and are in the process of writing to people who migrated from the area. In this manner, we have picked up at least 2 more patients which increases the potential significance of this cluster. In Mossyrock, Washington we learned of 6 patients in 3 families, all born between 1916 and 1920, and an additional unrelated patient born in 1940. Of possible significance, is the fact that a smallpox epidemic occurred in this area in 1924, and in the same year there were 10 infant deaths listed as being caused by measles. In the area around Wichita, Kansas we are in the process of studying a large series of patients and controls who were born in that area. We are concentrating on the sequence of infections and exposures which occurred before age 20, through questioning the mothers about the patients and controls, in a detailed manner. As would be expected, we are having difficulty finding enough patients and controls whose mothers are available for interview. We expect, however, to have enough cases for analysis within the next year.

Significance to biomedical research and the program of the Institute: Some aspect of geographic latitude has consistently been associated with the risk of multiple sclerosis both in the United States and throughout the world. The newer data on migrants substantiates the role of geographic latitude. To date these are the most important epidemiologic clues to the etiology of multiple sclerosis. The continuing search and investigation of well-defined migrant groups and special communities may provide further clues to the etiology of multiple sclerosis. The continuing search and investigation of well-defined migrant groups and special communities may provide further clues in understanding the factors contained in the broad concept of geographic latitude.

Proposed course: These studies will be continued and expanded through prevalence and case-control studies.

Honors and Awards: None

Publications:

Detels, R., Brody, J.A. and Edgar, A.H.: Multiple sclerosis among American, Japanese and Chinese migrants to California and Washington. J. Chron. Dis. 25:3-10, 1972.

Brody, J.A.: Comments on the epidemiology of multiple sclerosis and a possible virus etiology. Lancet, II:173, 1972.

Brody, J.A.: Epidemiologic and serologic data on multiple sclerosis and their possible significance. In Multiple Sclerosis: Immunology, Virology, and Ultrastructure. UCLA Forum in Medical Sciences, #16. Eds. F. Wolfgram, G.W. Ellison, J.G. Stevens and J.M. Andrews. Chap. 6, pp. 127-141. Academic Press, New York, 1972.

Serial No. NDS (CF) - 70 E 1838
1. Collaborative & Field Research
2. Epidemiology Branch
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Study of the incidence of Parkinson's disease among offspring of Parkinsonians

Previous Serial Number: Same

Principal Investigators: Roger Detels, M.D.
Jacob A. Brody, M.D.

Other Investigators: None

Cooperating Units: Church of Jesus Christ of Latter-day Saints,
Salt Lake City, Utah
The Genealogic Society,
Salt Lake City, Utah

Man Years:

Total:	3/16
Professional:	1/16
Other:	1/8

Project Description:

Objectives: To determine if genetic factors play a part in the etiology of Parkinson's disease by examining the incidence of disease among offspring of probands.

Methods employed: Fifty members of the Church of Jesus Christ of Latter-day Saints who died with the diagnosis of Parkinson's disease will be selected for review of death certificates in Utah between 1930 and 1940 and an additional 50 members who had a hospital diagnosis of Parkinson's disease between 1930 and 1940 selected from a review of hospital records from the major hospitals in the Salt Lake City area. A matched index control will be selected for each proband by taking the next death certificate or hospital record of an individual of the same sex whose age was within 5 years of the proband at the time of death or illness. Offspring of probands and index controls will be identified, a review of hospital records, of obituary notices, and a review of the records of the Genealogic Society and of the Church files of the Church of Jesus Christ of Latter-day Saints. Addresses of offspring will be obtained from the Church and questionnaires will be sent out to the offspring. Cases of Parkinson's disease will be confirmed when possible by physical examination or in the case of death through available medical records.

Major findings: None

Significance to biomedical research and the program of the Institute: In a chronic condition with late onset, such as Parkinson's disease, it is often difficult to determine the role of genetic factors. Members of the Church of Jesus Christ of Latter-day Saints maintain accurate, up-to-date genealogies. Thus, offspring of Parkinsonians can be located and the incidence of Parkinson's disease among them determined, providing a relatively simple method for determining the genetic factor in the etiology of Parkinson's disease.

Proposed course: See "Methods employed." Further data collection and analysis will be done by Dr. Detels, formerly with NINDS, presently with the University of California at Los Angeles.

Honors and Awards: None

Publications: None

Serial No. NDS (CF) - 70 E 1839
1. Collaborative & Field Research
2. Epidemiology Branch
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Studies on the relationship of chromosomal abnormalities and certain viral infections in mice. (Alternate title: Effects of viruses on mitosis and meiosis in the mouse.)

Previous Serial Number: Same

Principal Investigator: Lon R. White, M.D., DB, NICHD

Other Investigators: Harrie Anne Sutton, Biologist, DB, NICHD
George Nemo, Ph.D.
Jacob A. Brody, M.D.

Cooperating Unit: Developmental Biology Branch, NICHD

Man Years:

Total:	1/8
Professional:	1/16
Other:	1/16

Project Description:

Objectives: a. To localize within dividing mouse cells in vitro, particularly in relation to chromosomes, the site of residence of the infecting viral genome (of the minute virus of mice - MVM). b. To search for evidence of injury by virus to chromosomes of spermatogonia and somatic cells. c. To determine whether perinatal infection results in infection of the germ cells and, if so, to determine the duration of infection as well as the chromosomal and reproductive consequences of such infection.

Methods employed: The viruses under study are two strains of MVM. The animal used is the Cumberland View Farms C57BL/6 mouse. The viruses are "grown" and quantitated in tissue culture. Intracellular MVM genome (infectious or template DNA) is localized by hydroxyapatite column chromatographic demonstration of DNA - DNA hybridization using chromosomal DNA and radioisotope "tagged" MVM DNA. Chromosome studies are carried out on short-term spleen cell cultures as well as on cytologic preparations made directly from the spleen, testes, and bone marrow of infected and control mice. Viral antigen is localized using immunofluorescent and immunohistochemical methods.

Major findings: a. A "fate" study in mouse spleen cells using tritium-labeled virus preparations showed no difference in autoradiographic grain

localization between control and virus infected cultures. These were carried out with "tagged" viruses of relatively low infectivity and specific activity. b. Meiotic preparations from the testes of adult mice were studied 4 days, 1 week, 3, 6, and 12 weeks following MVM infection. The only notable observation was of a relative decrease in number of certain cell types, suggesting an effect on the course of spermiogenesis. c. Several animals initally infected in the perinatal period have produced normal litters.

Major improvements in methods involved with the propagation and quantitation of MVM have been accomplished. These have been used to produce viral "reagents" to be used in addition experiments. It was found that tritium labeling of viral DNA did not provide a sufficiently high specific activity to allow for autoradiographic localization of viral DNA in infected cells. An effort is being made to obtain high specific activity by labeling the DNA with I^{125} *in vitro*.

The DNA hybridization technique has been used to detect viral nucleic acid in whole cell DNA preparation from animals and from cells in culture. Current work is focused on the use of chromosomal DNA in order to determine if the viral genome may be integrated into or associated with the host DNA.

Significance to biomedical research and the program of the Institute: The role of viruses in the etiologies of chromosomal and developmental diseases is widely discussed, but negligibly investigated. The interaction of noncytolytic, non-oncogenic viruses with chromosomes and the spindle is also essentially undefined. The potential of these investigations is vast; basic information may be gained important to an understanding of human disease pathogenesis as well as to current concepts on the evolution of viruses and the basic nature of their interactions with dividing, differentiating cells.

Proposed course: To be continued at same level of effort.

Honors and Awards: None

Publications: None

Serial No. NDS (CF) - 70 E 1840
1. Collaborative & Field Research
2. Epidemiology Branch
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1971 through June 30, 1972

Project Title: Retinoblastoma, Clinical, Genetic and Psychometric Aspects

Previous Serial Number: Same

Principal Investigator: Roswell Eldridge, M.D.

Other Investigators: Kathy O'Meara
Jeffrey C. Allen, M.D.
David Kitchen, M.D.

Cooperating Units: John Hopkins Hospital, Baltimore, Maryland
Children's Hospital of D.C., Washington, D.C.
Department of Psychology, Downstate Medical Center
New York

Man Years:

Total:	1/2
Professional:	1/4
Other:	1/4

Project Description:

Objective: Only a few traits are said to be associated with increased intelligence. Such a correlation has been documented for torsion dystonia, the recessive type. Retinoblastoma is another condition for which such an association has been reported. Because we have reservations about choice of controls and value of data obtained in a handicapped population, we wished to evaluate this association in retinoblastoma ourselves.

Methods employed: Efforts were concentrated on testing the psychometric performance of affected individuals with family history who were sighted. Close unaffected relatives formed the control group.

Major findings: Twenty-three affected individuals and 23 control in 14 families have been evaluated by us. In addition, retrospective data from school groups testing wad obtained. We find that individuals affected with retinoblastoma and their unaffected relatives are unusually bright.

Significance to biomedical research and the program if the Institute: Among possible explanations for this observation are that members of bright families are on the average older at the time of child bearing because of longer schooling so that offspring are at greater risk for inheriting germ

cell mutation, or that brigher parents are more observant and that their affected offspring stand a better chance of survival and have access to more medical resources.

Proposed course: This Project is Terminated.

Honors and Awards. None

Publications: Eldridge, R., O'Meara, K., and Kitchin, D.: Superior Intelligence in Sighted Retinoblastoma Patients and Their Families. Journal of Medical Genetic, April 1972 (In Press)

Serial No. NDS (CF) - 70 E 1843
1. Collaborative & Field Research
2. Epidemiology Branch
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Causes of death among siblings of MS and ALS patients

Previous Serial Number: Same

Principal Investigator: Milton Koch, M.D.

Other Investigator: Jacob A. Brody, M.D.

Cooperating Unit: Department of Neurology, Duke University Medical Center
Department of Neurology, University of North Carolina Medical Center

Man Years:

Total:	3/16
Professional:	1/16
Other:	1/8

Project Description:

Objectives: Recently, we have developed serologic evidence that familial patterns of antibodies of MS patients differ significantly from controls. This suggests a familial process may be involved etiologically in MS. We wish to determine, therefore, if siblings of MS patients have unusual illnesses possibly related to immunologic defect. The diseases we are interested in include collagen diseases, thyroid diseases, and rheumatoid arthritis.

Methods employed: We will control the siblings of MS patients with the siblings of ALS patients. We are attempting to contact siblings of at least 44 ALS and 84 MS cases recently diagnosed in North Carolina. In addition, we have information on approximately 100 cases of ALS from North Carolina, who had this diagnosis on death certificates.

Major findings: Earlier attempts to locate siblings of 100 MS and 100 ALS patients from the autopsy file of Montifiore Hospital, New York City, have been unsuccessful.

Significance to biomedical research and the program of the Institute: There is no firm evidence that an autoimmune mechanism or a virus is the cause of MS. There is, however, accumulating suggestive evidence that this is the case. There is also evidence of altered serologic responses among siblings of MS patients. If we can document that the siblings of MS patients

have illnesses related to immunological mechanisms we would have valuable evidence that these mechanisms are involved in the etiology of MS.

Proposed course: Attempts using this new patient source are being made in hopes of getting better access to follow-up data on siblings.

Honors and Awards: None

Publications: None

Serial No. NDS (CF) - 70 E 1844
1. Collaborative & Field Research
2. Epidemiology Branch
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Amyotrophic lateral sclerosis in veterans

Previous Serial Number: Same

Principal Investigators: John Kurtzke, M.D.
Chief, Neurology Service, Veterans Administration Hospital, Washington, D.C.
Gilbert Beebe, M.D.
Director, Follow-up Agency, National Research Council of the National Academy of Sciences
James Norman, M.D.
National Research Council of the National Academy of Sciences
Virginia C. Karl, M.A.
Social Work Service, Veterans Administration Central Office, Washington, D.C.

Other Investigators: Jacob A. Brody, M.D.
Milton Koch, M.D.
Dwayne Reed, M.D.

Cooperating Units: Veterans Administration
National Research Council of the National Academy of Sciences

Man Years:

Total:	3/8
Professional:	1/4
Other:	1/8

Project Description:

Objectives: We will determine if there are distinctive epidemiologic features related to amyotrophic lateral sclerosis using the veteran population. We will also determine if service in Guam and the Mariana Islands was a risk factor in developing ALS. Life-table estimates will be constructed to clarify prognosis as to survival with ALS.

Methods employed: The methods paralleling those of the veterans study of multiple sclerosis will be used to obtain the "records sample." In addition, we will survey approximately 200 veterans with ALS and 200 controls with brain tumors using a detailed historical questionnaire. This population

will be called the "interview sample." Finally, a prognostic study will be constructed from Veterans Administration Hospital admissions of the 1957 - 1964 period for ALS and other motor neuron diseases. We directly participate in the "interview sample" phase of this contract (see Contract #PH 43-64-44, Task Order 62).

Major findings: A feasibility study of the extensive questionnaire was successful and we have now received approximately 50 questionnaires from patients and controls.

Significance to biomedical research and the program of the Institute: Regional differences and socioeconomic differences have been commented upon for amyotrophic lateral sclerosis in the United States. This study will determine for a large population if there are predisposing factors to this disease. Should we determine such factors, it would provide a valuable clue to the understanding, treatment and prevention of this disease. Prognosis for survival should also be clarified.

Proposed course: With our collaborators, we will collect all known veterans with ALS and try to detect possible predisposing factors. The contract commenced on July 1, 1972 and will run for 3 years.

Honors and Awards: None

Publications: None

Serial No. NDS (CF) - 70 E 1845
1. Collaborative & Field Research
2. Epidemiology Branch
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Natural history of Parkinson's disease and the effect of L-Dopa

Previous Serial Number: Same

Principal Investigator: Jacob A. Brody, M.D.

Other Investigator: Marjorie Matthews, R.N.

Cooperating Units: National Parkinson Foundation, Inc., Miami, Florida
Medical Care Insurance Corporation, Saskatchewan, Canada

Man Years:

Total:	5/16
Professional:	1/16
Other:	1/4

Project Description:

Objectives: To determine the long-term effects of L-Dopa on the natural history of Parkinson's disease and possible harmful effects of this medication over time.

Methods employed: While a case-control study would be the optimum way of determining the long-term effects of L-Dopa it would not be ethical at the present time to withhold the drug from a large number of cases. We have therefore been trying to identify populations in which there was sufficient information on a pre-L-Dopa cohort to determine a life table which could be compared with a cohort on L-Dopa. We have worked with the National Parkinson Foundation, Inc. in Miami and with the New York Hospital, but in each case, either follow-up has proven impossible or self-selection of patients in the pre-L-Dopa cohort made comparison with the post-L-Dopa cohort invalid. During the past year we have been attempting to determine if a valid pre- and post-L-Dopa cohort could be identified in Saskatchewan. The Province of Saskatchewan began a socialized medicine program in 1962, with the unique feature of having a central recording system. L-Dopa was not used until 1970. The overall population in one million with a relatively high proportion in older age groups. Thus, it might be possible to utilize this population for a suitable study.

Major findings: Previous efforts in the U.S. have not been successful (see above). We reviewed data available in Saskatchewan and have proposed a feasibility study.

Significance to biomedical research and the program of the Institute: It is not known whether L-Dopa provides only symptomatic relief of Parkinson's disease or whether it actually affects the pathologic process which produces this chronic illness. If indeed it does reverse the pathologic process, we would advance the understanding of the genesis of this disease. Since L-Dopa is a drug with known biohazards, we will be able to determine if these hazards shorten the life expectancy of treated patients or if they die of specific diseases such as cardiovascular disease or kidney disease which will permit us to focus medical attention on prevention of these specific complications among treated patients.

Proposed course: We are awaiting the outcome of the proposed feasibility study which was submitted by the University of Saskatchewan to the Canadian Research Council for funding.

Honors and Awards: None

Publications: None

Serial No. NDS (CF) - 71 E 1917
1. Collaborative & Field Research
2. Epidemiology Branch
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Twin study of multiple sclerosis: An epidemiologic inquiry

Previous Serial Number: None

Principal Investigators: A. Roger Bobowick, M.D.
Jacob A. Brody, M.D.

Other Investigators: John F. Kurtzke, M.D.
Veterans Administration Hospital, Washington, D.C.
Zdenek Hrubec, Sc.D.
Follow-up Agency, NAS-NRC
Marjorie Matthews, R.N.

Cooperating Units: Neurology Service, Veterans Administration Hospital, Washington, D.C.
Follow-up Agency, National Academy of Sciences, National Research Council

Man Years:

Total:	9/16
Professional:	1/16
Other:	1/2

Project Description:

Objectives: The abundant epidemiological literature of multiple sclerosis has indicated beyond reasonable doubt that critical environmental factors influence the rate of disease. Prevalence and migration studies strongly suggest that the environmental factor(s) is operant about the time of puberty. In order to examine the nature of the etiologic exposure in multiple sclerosis then, it is most prudent to concentrate on events in patients and controls with comparable early life histories in whom host factors are equalized. Twins discordant for multiple sclerosis at an age beyond prime risk of acquiring MS offer this opportunity for study.

Methods employed: The NAS-NRC twin registry has identified 23 pairs of twins one or both of whom has multiple sclerosis. This group has come from their population of 16,000 pairs of white male twins who are veterans of military service in World War II and born during the years 1917 to 1927. Information was collected concerning: 1) pertinent medical history and neurological exam, 2) an indepth epidemiologic interview concentrating on events prior to age 20, and 3) blood samples.

Major findings: Initial contact has been made with all of the twin pairs where possible. Nine twin pairs have been visited. Others could not be located, diagnosis could not be confirmed, or the patient refused.

Significance to biomedical research and the program of the Institute: Although exhaustive studies have been conducted to invoke specific etiologic factors in MS, no definitive precipitative circumstances have been identified perhaps because of the subtlety of these factors or the difficulties of suitably controlling for numerous unrelated circumstances. This novel application of the twin method of study offers an efficient technique for identifying these precipitating circumstances. Resolution of the cause and prevention of MS would be greatly enhanced by the identification of these precipitating factors.

Proposed course: Termination pending completion of analysis.

Honors and Awards: None

Publications: None

Serial No. NDS (CF) - 71 E 1918
1. Collaborative & Field Research
2. Epidemiology Branch
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Follow-up of the Cooperative Measles Vaccine Field Trial in four communities

Previous Serial Number: Same

Principal Investigators: Jacob A. Brody, M.D.
Anne H. Edgar

Other Investigators: Marion Dressler, M.D.
DeKalb County Health Department, Decatur, Georgia
Margaret I. Rathbun, M.D.
Monroe County Health Department, Rochester, New York
Edwin P. Isacson, M.D.
State University of New York School of Medicine
Russell E. Alexander, M.D.
University of Washington School of Medicine, Seattle
Philip S. Brachman, M.D.
National Communicable Disease Center, Atlanta

Cooperating Units: DeKalb County Health Department, Decatur, Georgia
Monroe County Health Department, Rochester, New York
State University of New York School of Medicine
University of Washington School of Medicine, Seattle
National Communicable Disease Center, Atlanta

Man Years:

Total:	3/16
Professional:	1/16
Other:	1/8

Project Description:

Objectives: To determine through contact with the participants in the killed measles vaccine field trial of the early 1960s, any possible sequelae to receipt of the vaccine.

Methods employed: Review of the addresses of the study population in four communities (DeKalb County, Rochester, Buffalo and Seattle) provided definite addresses for 46% of the 2091 participants plus possible location of an additional 17%. In order to determine the likelihood of success of follow-up, questionnaires requesting history of childhood diseases, other infections, non-allergic diseases accompanied by a rash, and hospitalizations

subsequent to receipt of the vaccine, were sent to the families of the study population in DeKalb County. To date, 92% of the definitely located families and 71% of the possibly located have responded. These same methods are to be extended to the participants in the field trial in the other three cities.

Major findings: To date the response to the study has been good but the small number of participants contacted allows no conclusions to be drawn.

Significance to biomedical research and the program of the Institute: The isolation of a measles-like virus from brains of individuals with subacute sclerosing panencephalitis has suggested a relationship between these two diseases. Epidemiologic study has further indicated that these patients may experience measles at an unusually early age. Evidence of high measles antibody titers in patients with multiple sclerosis has raised the possibility that measles might play an etiologic role in this disorder. In addition reports have been published on an atypical measles illness occurring in children who have received killed measles virus vaccine several years earlier. In light of the increasing interest in measles as an etiologic factor in disease, and the specific sequela reported to occur in those who have received the killed measles vaccine, the medical history of this study population subsequent to vaccination is of particular interest.

Proposed course: This project has been terminated.

Honors and Awards: None

Publications: None

Serial No. NDS (CF) - 71 E 1920
1. Collaborative & Field Research
2. Epidemiology Branch
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: A study of pregnancies among nurses; high risk pregnancies with possible viruses exposure

Previous Serial Number: Same

Principle Investigator: Estelle Kornhauser, R.N.

Other Investigator: Jacob A. Brody, M.D.
Marjorie Matthews, R.N.
Anne H. Edgar

Cooperating Unit: None

Man Years:

Total:	9/16
Professional:	1/16
Other:	1/2

Project Description:

Objectives: This project was developed to determine if nurses with frequent virus exposure have increased risk of abnormal pregnancy.

Methods employed: A questionnaire was designed using the usual backup history and pertinent questions relating to type of nursing service, year of pregnancy, normal or abnormal births, and all information relating to any type of exposure to a viral type disease. The questionnaire and letter of explanation was pre-tested in 1125 nurses registered in the Maryland and District of Columbia area.

Major findings: Of the 1125 questionnaires sent we have received 863 replies. To date there are approximately 1800 pregnancies including approximately 250 miscarriages, stillbirths, and abortions.

Significance to biomedical research and the program of the Institute: This study will determine if pregnant women working with people with infectious disease have an elevated risk of fetal damage.

Proposed course: The responses to date have been coded and are being tabulated for preliminary analysis. The results of this analysis will provide direction for expansion of the study to a larger number of respondents.

Honors and Awards: None

Publications: None

Serial No. NDS (CF) - 71 E 1921
1. Collaborative & Field Research
2. Epidemiology Branch
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Measles infection of mice: Effect of maternal immunity

Previous Serial Number: Same

Principle Investigators: Jacob A. Brody, M.D.
George Nemo, Ph.D.

Other Investigator: Lon R. White, M.D., DB, NICHD

Cooperating Unit: None

Man Years:

Total:	1/8
Professional:	1/16
Other:	1/16

Project Description:

Objectives: To develop an experimental animal model to study the pathogenesis of subacute sclerosing panencephalitis (SSPE).

Methods employed: Adult female mice previously immunized with measles virus will be bred and their offspring challenged with measles at various times after birth. Measles antibody levels will be determined periodically and a thorough analysis of mouse tissues for measles virus and measles-related antigens will be performed.

Major findings: Newborns of measles-immune mothers displayed a significant degree of passive immunity after challenge with measles virus. However we have thus far been unable to produce a chronic measles encephalitis in these survivors. We have taken representative survivors and prepared tissue culture from brains and other major organs. Cultures have been observed for measles cytopathology and tested for measles antigens employing fluorescent-antibody techniques. All tests have been negative. Certain groups of survivors are also being followed which received injections of live measles-infected vero cells. Other mice have received injections of cells obtained by trypsinizing the brains of measles-infected newborns. Several groups of survivors are being treated with cytoxan at various times after infection.

Significance to biomedical research and the program of the Institute: An agent which by most virological criteria resembles measles virus has been isolated from brains of SSPE patients. A papova-like virus has been visualized in electron micrographs of specially treated cell lines inoculated with SSPE brain material suggesting the possibility of a dual infection. The exact role these agents play in the pathogenicity of the disease is unclear. An experimental animal model may aid considerably in elucidating the mechanism.

Proposed course: Newborns which have survived measles inoculations will be observed continually for unusual signs. We are currently inoculating groups of survivors with other viral agents in hopes of triggering an SSPE-like syndrome.

Honors and Awards: None

Publications: None

Serial No. NDS (CF) - 71 E 1922
1. Collaborative & Field Research
2. Epidemiology Branch
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Search for a negative DNA strand in cells infected with the minute virus of mice (MVM)

Previous Serial Number: Same

Principle Investigator: Lon R. White, M.D., DB, NICHD

Other Investigators: Harrie Anne Sutton, Biologist, DB, NICHD
George Nemo, Ph.D.
Jacob A. Brody, M.D.

Cooperating Unit: Developmental Biology Branch, NICHD (HD DB - 15)

Man Years:

Total:	1/8
Professional:	1/16
Other:	1/16

Project Description:

Objectives: a. To demonstrate a "template" DNA (negative strand) complementary to the single-stranded DNA (positive strand) of the MVM virion. b. To determine if the negative strand is associated with the cell's chromosomal DNA. c. To investigate the kinetics of negative strand synthesis relative to cellular DNA replication, cellular division, virus infection, and virus production. d. To utilize this method to detect a latent or cryptic viral infection in the cells of experimental animals and in tissue culture. e. To characterize the virion DNA, the "negative strand," and replicative forms of MVM DNA.

Methods employed: These studies utilize radioisotope "tagged" viral DNA. Annealing with "cold" DNA from virus infected cells is demonstrated by liquid scintillation detection of double- and single-stranded DNA in fractions eluted from a hydroxyapatite column.

Major findings: Contrary to expectations, DNA extracted from purified, concentrated virus preparations is inhomogeneous and partially double-stranded. A significant amount of additional duplex formation results when the initially single-stranded DNA is allowed to "self-anneal." That these sequences are truly viral and not from the host cell is shown by the failure of added mouse or rat DNA to influence the rate or extent of annealing. Denaturation and reannealing kinetics of the initially double-stranded

suggests the presence of one or more loop or hairpin molecular configurations. Nucleic acid hybridization reactions using total cell DNA and a "stripped" (all double-stranded parts removed) viral DNA "probe," has now been used to detect complementary sequences in DNA from both acutely infected L cells (tissue culture) and newborn mice. Negative results were found with normal mouse (3 strains), rat, and guinea pig DNA, as well as with DNA from 2 tissue culture lines. Unexpectedly, positive results were found with DNA from a leukemic rat. Current work is focused on the utilization of this method to detect a latent or persistent parvovirus infection in animals and tissue culture.

Significance to biomedical research and the program of the Institute: Characterization of virus-cell interactions using a single-stranded DNA virus (parvovirus group, of which MVM is representative) is expected to advance our understanding of the mechanisms by which a virus infection might alter the genetic potential of the host cell (either a somatic or germ cell) as well as of mechanisms involved with the establishment of latent and chronic viral infections. Congenital, chronic, or latent virus infections may be involved with the etiologies of certain reproductive, developmental, and chronic diseases whose causes are now unknown. This experimental approach may prove to be directly applicable to a search for a parvovirus DNA in the cells in animals and persons with such diseases.

Proposed course: To be continued at an increased level of effort.

Honors and Awards: None

Publications: None

Serial No. NDS (CF) - 71 E 1923
1. Collaborative & Field Research
2. Epidemiology Branch
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Development of *in vitro* cellular immune tests for viral antigens

Previous Serial Number: Same

Principal Investigators: George Nemo, Ph.D.
Jacob A. Brody, M.D.

Other Investigator: None

Cooperating Unit: None

Man Years:

Total:	1/8
Professional:	1/16
Other:	1/16

Project Description:

Objectives: To develop lymphocyte transformation assays using human lymphocytes and highly purified measles virus and vaccinia virus as test antigens.

Methods employed: We have developed purification procedures for various strains of measles virus (e.g. SSPE, low and high passage Edmonston) and vaccinia virus which includes differential centrifugation, ammonium sulfate precipitation and density gradient techniques. Lymphocytes are cultured employing standard procedures. Purified viral preparations either live or UV-irradiated are added and serve as viral antigens. Tritiated thymidine is added 5 hours prior to harvest and uptake of the isotope is measured by liquid scintillation spectrometry.

Major findings: We have been successful in our attempts to develop an *in vitro* lymphocyte transformation assay in humans using highly purified preparations of measles and vaccinia virus. Our experiments have shown that the peak of lymphocyte stimulating activity for both measles and vaccinia occurs on day 7. This agrees well with other specific antigens such as PPD and candida which also peak on day 7. Density gradient fractions of measles virus were capable of stimulating lymphocytes with the peak of stimulatory activity coinciding with the peak of virus infectivity. UV-irradiated preparations of measles and vaccinia are also capable of stimulating human lymphocytes.

Significance to biomedical research and the program of the Institute: MS patients have been shown to possess elevated measles antibody titers when compared to appropriate controls. Furthermore, the recent finding that spinal fluid of MS patients contains antibody to vaccinia virus has prompted the inclusion of purified vaccinia virus as a test antigen in our cellular assay. We are particularly interested in measuring the cellular immune reactivity of MS patients and controls to purified measles and vaccinia antigens in hopes that the relationship of these agents to MS may be determined.

Proposed course: After the basic work is completed we intend to use the test to measure lymphocyte responsiveness to these particular antigens in large groups of MS patients and controls.

Honors and Awards: None

Publications: None

Serial No. NDS (CF) - 71 E 1924
1. Collaborative & Field Research
2. Epidemiology Branch
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Biochemical Studies in Torsion Dystonia

Previous Serial Number: Same

Principal Investigator: Roswell Eldridge, M.D.

Other Investigators: G. Frederick Wooten, M.D.

Cooperating Unit: Laboratory of Clinical Science, NIMH

Man Years:

Total:	3/8
Professional:	1/8
Other:	1/4

Project Description:

Objectives: Torsion dystonia has been shown recently to consist of at least two hereditary conditions as well as an acquired form. Through appropriate biochemical investigation it should be possible to determine the precise biochemical defect (or abnormal gene product) in the recessive form of torsion dystonia.

Methods employed: Plasma has been collected from affected patients, quick frozen, and assayed for various catecholamine levels by Dr. Wooten.

Major findings: Observations in 40 patients with torsion dystonia indicated that the enzyme dopamine beta hydroxylase (DBH), which governs the conversion of dopamine to noradrenalin, is present in abnormally high levels in individuals with the dominant form of torsion dystonia. This work was carried out by Dr. Frederick Wooten in the laboratory of Dr. Julius Axelrod.

Significance to biomedical research and the program of the Institute: Demonstration of the molecular lesion in one of the torsion dystonias would be a noteworthy achievment in the quest for inborn errors of metabolism. In addition, important information may develop regarding biochemistry and physiology of movement control. Specific treatment and pre-clinical detection useful in the genetic counselling of these families

May be more immediate rewards. In addition, because of the recognized association of increased intelligence and the recessive form of dystonia, we stand to gain information regarding the development of intelligence.

Proposed course: Continued investigation of patients from the genetics registry. Addition of more sophisticated biochemical techniques as they become available.

Honors and Awards: None

Publications:

Eldridge, R.: The Torsion Dystonias: Literature Review: Genetic and Clinical Studies. In The Torsion Dystonias (Dystonia Musculorum Deformans). Editor, Roswell Eldridge. Neurology, suppl. 20:11, part 2, pp. 1-78, November 1970.

Chase, T.N.: Biochemical and Pharmacologic Studies of Dystonia. In The Torsion Dystonias (Dystonia Musculorum Deformans). Editor, Roswell Eldridge. Neurology, suppl. 20:11, part 2, November 1970.

Eldridge, R., Harlan, A., Cooper, I.S., and Ricklan, M.: Superior Intelligence in Recessively Inherited Torsion Dystonia. The Lancet, I:7637, pp. 65-67, 1970.

Wooten, F., Eldridge, R., Axelrod, J., and Stern, R.: Elevated Plasma Dopamine Beta Hydroxylase Activity in Autosomal Dominant Torsion Dystonia. New England Journal of Medicine, 288:6, pp. 284-287, 1973.

Wooten, F., Eldridge, R., Gordon, E.K., and Axelrod, J.: Studies of Catecholamine Metabolism in Two Hereditary Forms of Dystonia. (In Press)

Serial No. NDS (CF) - 71 E 1925
1. Collaborative & Field Research
2. Epidemiology Branch
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1. 1972 through June 30, 1973

Project Title: Intelligence in Israeli Patients with Torsion Dystonia

Previous Serial Number: Same

Principal Investigators: Roswell Eldridge, M.D.

Other Investigators: Morris Gross, PhD.
Richard Goodman, M.D.
Thelma Koerber

Cooperating Units: Department of Education
Herbert H. Lehman College
Bronx, New York

Man Years:

Total:	5/16
Professional:	1/4
Other:	1/16

Project Description:

Objectives: The recessive form of torsion dystonia has recently been shown to be associated with increased intelligence. However, the series which this information is based was small and individuals were scattered over a large area of the northeastern part of the United States. In Israel, where we suspect there to be over 20 patients with the recessive form of torsion dystonia, a more homogeneous testing situation prevails so that results should be more meaningful. If this association is true the implications are dramatic so that confirmation to this Israeli study is clearly in order.

Methods employed: Bona fide cases of torsion dystonia in Israel would be selected by the principal investigator on the basis of personal examination. Psychometric data would be obtained in a retrospective manner through the Israeli Department of Education and similar data would be obtained for unaffected sibs. A comparison would then be made of the performance in these two groups.

Major findings: Although we have known for several years of the presence of over 20 cases of torsion dystonia in Israel we have not been

able to arrange for visitation. Neurologists, psychologists, and educators in Israel have been alerted and indicated their interest in participating in this study.

Significance to biomedical research and the program of the Institute: The positive association between torsion dystonia and intelligence would suggest that the chemical abnormality producing dystonia may also enhance intellectual development. Elucidation of this basic chemical abnormality would suggest a method for enhancing intellectual development, particularly those from the retarded population.

Proposed course: Several weeks in Israel would be necessary to make the necessary home visits and arrange for the collection of appropriate psychometric data but travel funds have not been available for over the past 3 years.

Honors and Awards: None

Publications: Eldridge, R., Harlan, A., Cooper, I.S. and Ricklan, M.: Superior Intelligence in Recessively Inherited Torsion Dystonia. The Lancet. I:7637, pp. 65-67, 1970.

Serial No. NDS (CF) - 71 E 1926
1. Collaborative & Field Research
2. Epidemiology Branch
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 20, 1973

Project Title: The Difficulty of Diagnosing Acoustic Neuroma in Young Patients

Previous Serial Number: Same

Principal Investigator: Roswell Eldridge, M.D.

Other Investigators: Thelma Koerber

Cooperating Units: Department of Neurosurgery
Columbia Presbyterian Hospital
New York

Man Years:

Total:	1/4
Professional:	1/8
Other:	1/8

Project Description:

Objectives: To document and publicize the difficulty of diagnosis when acoustic neuroma occurs in young patients.

During a course of clinical and genetic study of acoustic neuroma in young individuals we have been impressed with the unusual length of time and unusual number of specialists consulted before diagnosis of acoustic neuroma became a consideration. This period was often one of unusual unnecessary stress and expense to the patient and his family and often resulted in loss of valuable time in many instances.

Methods employed: Patients who have early onset of acoustic neuroma were questioned in detail regarding physicians consulted and diagnostic procedures employed.

Major findings: In general, the younger the individual when first symptomatic, the longer the period before diagnosis, the greater the number of physicians consulted and the more expense to the family.

Significance to biomedical research and the program of the Institute: Such documentation should impress the medical community about the resistance of this condition in young individuals thereby reducing time, expense and turmoil to patient, family, and physician when a new case develops. Through such publicity it is possible that our acoustic neuroma registry will be expanded.

Proposed course: Documentation of similar experience in the remainder of our acoustic neuroma patients. Publication of results.

Honors and Awards: Presentation at Middle Atlantic Neurosurgery Society

Publications: Allen, J., and Eldridge, R.: Genetic Study of Early Onset Acoustic Neuroma. Proceedings of the Fifth Conference of the Clinical Delineating of Birth Defects, June 1972. The National Foundation-March of Dimes. (In Press)

Serial No. NDS (CF) - 71 E 1927
1. Collaborative & Field Research
2. Epidemiology Branch
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Family Studies, Including Histopathology of Cafe-au-lait Spots, in Early Onset Acoustic Neuroma

Previous Serial Number: Same

Principal Investigator: Robert Stern, M.D.

Other Investigators: Roswell Eldridge, M.D.
Martin Mihm, M.D.

Cooperating Unit: Department of Dermatology
Harvard Medical School

Man Years:

Total:	1/4
Professional:	1/8
Other:	1/8

Project Description:

Objectives: To clarify the relationship of classical "peripheral" neurofibromatosis to early onset hereditary bilateral acoustic neuroma by histological study of cafe-au-lait spots in each disorder. To evaluate these findings as a possible diagnostic marker for the former disease.

Methods employed: Obtain skin punch biopsies from individuals with, or at risk for, early onset bilateral hereditary acoustic neuroma. Examine the histopathology of these tissue specimens comparing them to the histologically distinct characteristics of cafe-au-lait spots found in "normal" individuals and those found in neurocutaneous syndromes.

Major findings: Pending

Significance to biomedical research and the program of the Institute: Benedict and Johnson have established that both the melanosome counts and characteristics of individual melanosomes are distinct in neurofibromatosis cafe-au-lait spots. Thus, through the study of the histopathology of cafe-au-lait spots, it may be possible to define the relationship between hereditary bilateral acoustic neuroma and

neurofibromatosis and also provide a genetic marker for hereditary acoustic neuroma. This latter information would be of great aid for purposes of genetic counselling and indicating those relatives that require maximum surveillance.

Proposed course: Proceed with investigators as outlined under "methods employed" as soon as clearance for biopsies is obtained from NINDS medical clearance committee.

Honors and Awards: None

Publications: None

Serial No. NDS (CF) - 71 E 1928
1. Collaborative & Field Research
2. Epidemiology Branch
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Biochemical and Chromosome Studies in Retinoblastoma

Previous Serial Number: Same

Principal Investigator: Roswell Eldridge, M.D.

Other Investigators: Frederick Wooten, M.D.
David Kitchen, M.D.
Robert Stern, M.D.

Cooperating Units: Eye Institute
Columbia Presbyterian Hospital
New York City
Laboratory of Clinical Pharmacology, NIMH

Man Years:

Total:	1/8
Professional:	1/16
Other:	1/16

Project Description:

Objectives: Retinoblastoma, an hereditary tumor of the eye, affects children in the first year or two of life. Early diagnosis is critical and can lead to life and sight saving therapy. Diagnosis now depends on complete retinal examination of at risk individuals as does evaluation of therapy. A screening test involving a specific tumor metabolite in blood or urine is needed to determine newborns who carry the retinoblastoma gene and in treated youngsters to evaluate if therapy has been adequate. Neuroblastoma, a nervous system tumor sharing same histologic similarities with retinoblastoma produces elevation in dopamine beta hydroxylase activity in plasma of many instances. We are studying dopamine beta hydroxylase to determine if it is elevated in retinoblastoma as well.

Methods employed: Treated and untreated patients and their sibs attending the Eye Institute comprise the sample population. Infants and children with other eye disorders comprise the control population. Blood samples are obtained in the morning, red cells are spun down and the plasma is frozen. DBH is determined in the laboratory of Dr. Julius Axelrod.

Major findings: Approximately 15 patients have been studied to date; although several seem to have high levels, thorough analysis is not yet completed.

Significance to biomedical research and the program of the Institute: If patients with active retinoblastoma have significantly higher DBH a valuable tool for early detection and monitoring therapy will be available. Also, the confusing genetics of retinoblastoma may be clarified.

Proposed course: Complete DBH screening of patients, sibs and controls.

Honors and Awards: None

Publications: None

Serial No. NDS (CF) - 71 E 1929
1. Collaborative & Field Research
2. Epidemiology Branch
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1971 through June 30, 1972

Project Title: Genetic Study of an Irish Isolate in South Carolina

Previous Serial Number: Same

Principal Investigator: Roswell Eldridge, M.D.

Other Investigators: Jeffrey C. Allen, M.D.
Kathy O'Meara
Charles Still, M.D.

Cooperating Unit: Department of Neurology,
University of South Carolina
Columbia, South Carolina

Man Years:

Total:	1/8
Professional:	1/16
Other:	1/16

Project Description:

Objectives: The Irish Travellers of Murphy Village, South Carolina are a unique Catholic isolate whose exact origins are unknown. The present population is approximately 1,100 of which 420 are under 20 years of age. The men receive income from spraying barns and laying linoleum. They secure their business by travelling in small trucks throughout the eastern United States.

The Village is of particular interest to us because of the apparent high rate of inbreeding which predisposes autosomal recessive traits.

Methods employed: At present the need is for precise demographic data such as population break down by sex and age. Patterns and family size would be of interest.

Genetic information may be best secured through conversation with responsible elders in the Village. Undoubtedly the group traces from a small founding population and precise information about this as well as information about members added to the group in recent years would be important.

The genetic relationship of this population to the present day Irish might be established by noting the gene frequencies with ABO, Rh and other accessible

footmarkers.

Medical genetic studies most certainly focus on existing recognized familial problems such as mental retardation and skin disease. In addition, a comprehensive medical evaluation of the community and historical account of disorders in earlier generations would be worthwhile. Screening for newborns looking for recognized metabolic and chromosomal abnormalities might be productive.

Major findings: Preliminary information has already been secured regarding the background, demography, economy, social organization, education, medical problems and genetics. Several familial traits are present among the Villagers suggesting that a comprehensive survey would reveal other genetic traits.

Significance to biomedical research and the program of the Institute: Studies of such isolates have been unusually productive in the delineation and understanding of genetic entities. Example include Victor McKusick's study of the Amish and Carl Witkop's study of the Wiesorts.

Proposed course: This Project Is Terminated

Honors and Awards: None

Publications: None

Serial No. NDS (CF) - 71 E 1930
1. Collaborative & Field Research
2. Epidemiology Branch
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Von Hippel-Lindau's Syndrome - Clinical, Genetic and Biochemical Aspects

Previous Serial Number: Same

Principal Investigator: Roswell Eldridge, M.D.

Other Investigators: Robert S. Stern, M.D.
Thelma Koerber

Cooperating Units: Department of Neurology
Johns Hopkins Hospital
Columbia Presbyterian Medical Center
New York

Man Years:

Total:	1/4
Professional:	1/8
Other:	1/8

Project Description:

Objectives: In 1911 Eugen von Hippel described a number of patients with retinal angiomatosis. In 1926 Arvid Lindau described a number of patients, who in addition to the retinal angiomatosis, had such intracranial lesions such as cerebellar cysts and cerebellar medullary angioblastic tumors as well as pancreatic cysts, renal cysts and adrenal tumors. The familial nature of this syndrome has since been noted by numerous authors.

What has impressed us is the infrequency with which these tumors are bilateral. In contrast, in other hereditary traits associated with neoplasm of paired structures the involvement is generally bilateral. Is the tendency to unilateral involvement in the Von Hippel-Lindau's syndrome real and, if so, does it reflect a different etiologic mechanism than seen in the other hereditary neoplasms? Or is the unilateral involvement only apparent, reflecting failure to scrutinize the presumably healthy member of the paired organs?

Methods employed: The propositi with Von Hippel's syndrome will be ascertained through neurologic departments of selected medical centers including the Armed Forces Institute of Pathology. Contact with

individuals and their families will be made through their personal physician, and home visits will be arranged for physical examination and detailed history.

Major findings: None

Significance to biomedical research and the program of the Institute: The first step will be to document whether or not this syndrome tends to be unilateral in its involvement. Later more sophisticated laboratory studies can be undertaken to determine more closely the mechanism of abnormal gene action which produces the neoplasias.

Proposed course: See "Methods employed".

Honors and Awards: None

Publications: None

Serial No. NDS (CF) - 71 E 1931
1. Collaborative & Field Research
2. Epidemiology Branch
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1971 through June 30, 1972

Project Title: Genetic Study of Population Isolates in Maine

Previous Serial Number: Same

Principal Investigators: Jeffrey Allen, M.D.
Roswell Eldridge, M.D.

Other Investigators: Morris Lambdin, M.D.
Ellsworth, Maine
Thomas Roderick, M.D.
Jackson Laboratory
Bar Harbor, Maine

Cooperating Units: Division of Public Health Nursing,
State of Maine Dept. of Health & Welfare
Maine Genetics Counselling Center
Ellsworth, Maine
Maine Medical Association; Lincoln, Sagadahoc,
Washington, Waldo and Hancock Counties

Man Years:

Total:	5/16
Professional:	1/16
Other:	1/4

Project Description:

Objectives: To investigate numerous genetic subisolates along the coast of Maine for evidence of hereditary neurologic disorders.

Methods employed: A preliminary trip was made to establish contacts and examine the feasibility of the study. Thereafter an introductory letter and brief questionnaire was sent to every registered physician and public health nurse in the five major coastal counties. The questionnaire was designed to provide information on the awareness of any unusual familial disorders; diagnosed and undiagnosed, which are prevalent in remote areas of the state, especially on some of the offshore islands.

If feasible, demographic and historical data will be collected from State, County and local sources to corroborate the prevalence of genetic isolation.

Patients and their families will be visited and an appropriate referral will be made to local diagnostic facilities.

Major findings: So far 60 of 119 (58 percent) questionnaires have been returned. Several interesting leads exist.

Significance to biomedical research and the program of the Institute: Maine's rugged eastern coast contains innumerable islands of varying sizes. Several of these have been inhabited for several generations and populations vary from 50 to 200 persons. Inbreeding is commonly practiced. On several islands, most of the inhabitants have only two or three different surnames. Because of the remoteness of the islands inhabitants have not availed themselves of the usual medical facilities. They are visited rather infrequently by a public health nurse or physician. The likelihood of finding recessive hereditary disorders is high in these populations.

Proposed course: This Project Is Terminated

Honors and Awards: None

Publications: None

Serial No. NDS (CF) - 72 E 1977
1. Collaborative & Field Research
2. Epidemiology Branch
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Selected Genetic and Clinical Aspects of Huntington's Disease

Previous Serial Number: Same

Principal Investigator: Robert Stern, M.D.

Other Investigators: Roswell Eldridge, M.D.
Marjorie Guthrie
Thelma Koerber

Cooperating Unit: Committee to Combat Huntington's Disease

Man Years:

Total:	3/8
Professional:	1/4
Other:	1/8

Project Description:

Objectives: To explore the attitudes of individuals with Huntington's disease and their relatives towards the disease, and to determine the sources of information about the disease, family planning, and genetic screening tests.

Methods employed: Questionnaires were sent by the Committee to Combat Huntington's Disease to 5,000 of their members.

Major findings: Analysis of more than one thousand responses has indicated the chief concern is with the physical aspects of this disease rather then mental, social, or genetic aspects. In addition, this group places reliance for information on a voluntary lay organization, the Committee to Combat Huntington's Disease, more than all medical sources combined. Awareness of the fifty percent transmission risk in Huntington's disease was significantly greater among those indicating Committee to Combat Huntington's Disease as the best source of information compared to those indicating family physician or relatives. The majority of respondents indicated that being at risk for Huntington's disease would result in their wanting fewer children. Eighty percent of at risk

individuals expressed an interest in a screening test.

Significance to biomedical research and the program of the Institute: Previous studies of genetic counselling and opinions of at risk or carrier individuals to their hereditary disease have been based on individuals who have had exposure to genetic counselling and other specialized services. This study is unique because the large number of respondents representing a relatively heterogeneous population were asked their opinions independent of any connection with such a program. As a result, the degree of concern varied among these individuals. Knowledge of these concerns should be of interest to genetic counsellors and others involved in the care of Huntington's disease families. As increasing emphasis is placed on the development of presymptomatic screening tests for individuals at risk for inherited diseases, this study provided information on the attitudes towards such tests of individuals who are both possible carriers of the defective gene and at the same time at risk for the disorder.

Lay groups such as CCHD can provide an efficient means for disseminating information and hope to patients and families confronted with one of the most distressing hereditary diseases.

Proposed course: In addition to completion of the analysis of responses already received, distribution of similar questionnaires to Huntington's families ascertained through state registries is anticipated. Comparison of responses from such a population provide a basis for comparison of attitudes and information about disease among groups being furnished information from separate sources.

Honors and Awards: None

Publications: None

Serial No. NDS (CF) - 72 E 1978
1. Collaborative & Field Research
2. Epidemiology Branch
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Geographic distribution of neurologic diseases within the United States

Previous Serial Number: None

Principal Investigator: Dwayne Reed, M.D.

Other Investigators: Anne H. Edgar
Marjorie Matthew, R.N.
Jonas Ellenberg, Ph.D.
Office of Biometry, NINDS
Alan J. Talbert
Office of Biometry, NINDS
Robert Buechley, Ph.D.
Environmental Protection Agency

Cooperating Units: Section on Mathematics Statistics, Office of Biometry, NINDS
Division of Health Effects Research, Environmental Protection Agency, Research Triangle Park, N.C.

Man Years:

Total:	9/16
Professional:	1/16
Other:	1/2

Project Description:

Objectives: To conduct a geographic analysis of the association of a broad variety of environmental variables with death rates for cerebrovascular disease (CVD) and multiple sclerosis (MS) and to analyze the interaction of environmental variables and the mortality rates to establish a perspective by which to evaluate clues for further hypothesis testing.

Methods employed: This project is being conducted through the joint efforts of the Epidemiology and Biometry personnel. The first step has been to obtain statistical data concerning mortality rates for CVD and MS and a variety of environmental variables including measures of demography, socio-economic levels, sanitation, pollution, social disruption and various aspects of climate. Standard metropolitan statistical areas (SMSA) were chosen as the geographic unit for analysis as they have populations large enough to have stable death rates and yet are not so large as to include wide variations in

the environmental factors. The statistical profiles of the study units have been tabulated and put on computer tapes for analysis.

Major findings: Correlation and stepwise multiple regression analyses have been completed for age-adjusted mortality rates for both MS and CVD in 200 SMSA's. For MS the results were consistent with earlier studies. The highest correlations were seen with environmental factors which were related to latitude; including geographic location, temperature, and amount of daily sunshine. No meaningful associations were found with measures of crowding, socioeconomic levels, sanitation, pollution or social disruption.

For CVD the findings were also similar to earlier studies. Geographic location in the south eastern United States and some measures of socioeconomic status had the highest correlations with high stroke death rates.

Significance to biomedical research and the program of the Institute: Death rates for CVD and MS have well known geographic distribution patterns within the continental United States. These patterns have been consistent and have stimulated searches for variables which have the same geographic distribution as the mortality rates. To date only isolated variables have been examined to explain the geographic patterns and no broad overview has been attempted. An understanding of the interrelation of the variables and the mortality rates will help establish a perspective by which to evaluate the role of these environmental variables as well as suggest clues for further analysis.

Proposed course: As no new hypotheses were generated from these analyses this project will be terminated.

Honors and Awards: None

Publications: None

Serial No. NDS (CF) - 72 E 1979
1. Collaborative & Field Research
2. Epidemiology Branch
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Studies on the consequences of viral infection in early life

Previous Serial Number: Same

Principal Investigator: Lon R. White, M.D., DB, NICHD

Other Investigators: Dwayne Reed, M.D.
Harrie Anne Sutton, Biologist, DB, NICHD
Jacob A. Brody, M.D.

Cooperating Unit: Developmental Biology Branch, NICHD (HD DB - 20)

Man Years:

Total:	1/8
Professional:	1/16
Other:	1/16

Project Description:

Objectives: To investigate immune responses and neurologic development in children known or suspected of having contracted certain virus infections during infancy or early childhood. The initial studies have focused on infection with "natural" measles during the first year of life.

Methods employed: A population of approximately 100 children in and around Little Rock, Arkansas, known to have had measles before one year of age were identified with the assistance of a local hospital physician and public health officers. To test the feasibility of an intensive, longitudinal study of this cohort and control children, an attempt was made to contact, evaluate and obtain serum from several "case" and "control" children. Sera were tested for anti-measles antibody by hemagglutination inhibition.

Major findings: No "control" children could be identified. Five "cases" were examined and were considered neurologically normal although their antibody titers were slightly elevated above expected levels.

Significance to biomedical research and the program of the Institute: Arbovirus infections involving the CNS and occurring in infancy or early childhood are known to be associated with signs of minimal brain damage as the children near school age. This occurs even though the original illness was mild and followed by apparently complete recovery. Subclinical CNS involvement with the common childhood illnesses (measles, mumps, herpes

simplex, varicella, roseola infantum) is frequent; the neurologic sequelae of these illnesses are undefined but may be of considerable importance. This study represents an attempt to define the role of such early infections in the causation of minimal brain damage and related neurologic abnormalities.

Proposed course: This project has been terminated.

Honors and Awards: None

Publications: None

Serial No. NDS (CF) - 72 E 1980
1. Collaborative & Field Research
2. Epidemiology Branch
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Development and application of methods for the study of the minute virus of mice (MVM)

Previous Serial Number: Same

Principal Investigator: Lon R. White, M.D., DB, NICHD

Other Investigators: Harrie Anne Sutton, Biologist, DB, NICHD
George Nemo, Ph.D.
M.D. Hoggan, Ph.D., LVD, NIAID

Cooperating Units: Developmental Biology Branch, NICHD (HD DB - 16)
Laboratory of Viral Diseases, NIAID

Man Years:

Total:	1/2
Professional:	1/4
Other:	1/4

Project Description:

Objectives: To develop improved methods for the propagation, purification, and quantitation of the virus and its nuclei acid. To investigate basic biological, biochemical, and structural properties of the virus and its DNA in order to develop techniques to investigate its persistence in congenitally and chronically infected experimental animals.

Methods employed: The agent is propagated in tissue culture and detected by cytopathic effect or antigen production. Its DNA is radioisotope "tagged" during replication. It is purified by differential centrifugation and by chemical means. The virus, its proteins, and its DNA are characterized by velocity sedimentation in sucrose gradients, electron micrographic appearance, salt and thermal elution characteristics from hydroxyapatite columns, and polyacrylamide gel electrophoresis.

Major findings: Previous reports have described the growth of MVM in only two cell lines, both primary, with CPE in only one of these. We have extended this to 4 continuous cell lines, 2 of which show CPE. We have shown that whereas maximum virus production demands that the host cell divide, infection can occur in non-dividing cells and the virus can remain "latent" without apparent injury to the cell until it divides. In addition, we have established chronic infection in four cell lines, two of which have been

producing virus in high concentration continuously for 2 years. We have accomplished a 100-1000 x improvement in yield of virus and hemagglutinating antigen and have shown that purification by isopycnic ultracentrifugation is associated with moderate virus disruption and loss of infectivity. Additional studies have revealed no significant loss of hemagglutination or infectivity titers with 3 cycles of freeze-thawing or with heating at 56°C for up to 90 minutes. UV inactivation progresses in approximately one $\log_{10}$ decrements per second under standard conditions. The in vitro infectivity titration method has been improved from a rather unreliable one-month procedure (which works poorly with "wild" strains of MVM) to a sharply reproducible, reliable one-week procedure usable with all strains. The nuclei acid, extracted by NaOH treatment and by a pronase-phenol-chloroform method, has been found to be extremely labile and "sticky," adsorbing to plastics, millipore filters, and dialysis tubing. Results of hydroxyapatite chromatography suggest that it may have a "snap back" area where it is double-stranded, that some virions may contain some "negative" strand DNA, and that the DNA may be packaged in segments. A method has been developed for the demonstration of viral DNA in infected cells and are being used to study the course of MVM infection in vitro and in vivo. The virus has at least 3 proteins, whose molecular weights are between 50,000 and 80,000, and whose relative concentrations most closely resemble the pattern observed with a bovine parvovirus. Differences in relative concentration have been observed with cultivation in different cell lines; these are currently under study.

Significance to biomedical research and the program of the Institute: MVM is representative of a group of small, single-stranded DNA viruses (the parvoviruses) which are characterized by ubiquity, generally low virulence and cytopathogenicity, an apparent preference for dividing cells, and a high frequency of transplacental infections. The study of this agent is expected to lead to a better understanding of mechanisms and the possible role of viruses in the causation of certain genetic, reproductive, and developmental abnormalities. MVM has been relatively unstudied, presumable because of technical problems. The improved methods which we have developed, and the basic information obtained by the application of these methods now make such studies feasible.

Proposed course: To be continued at present level of effort.

Honors and Awards: None

Publications: None

Serial No. NDS (CF) - 73 E 2045
1. Collaborative & Field Research
2. Epidemiology Branch
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Epidemiology of polymyositis in children

Previous Serial Number: None

Principal Investigators: Milton Koch, M.D.
Marjorie Matthews, R.N.
Jacob A. Brody, M.D.

Other Investigator: None

Cooperating Units: Various neurologic and pediatric centers

Man Years:

Total:	1 1/2
Professional:	1/2
Other:	1

Project Description:

Objectives: Polymyositis in children under 16 years of age will comprise the first phase of this investigation. Recently, several reports have indicated the presence of myxovirus-like particles in muscle biopsy of affected patients. We wish to determine, by administration of a questionnaire to a case-control population, if we can detect distinctive antecedent factors which may be predisposing or causative in this disease.

Methods employed: We have contacted several medical centers requesting the names of patients with documented diagnosis of polymyositis, under age 16, diagnosed within the last 5 years. The parents of these patients are contacted by us to obtain permission to administer a questionnaire, and to find a suitable control. Then, this questionnaire is administered both to the parents of the patients and the controls. Blood is drawn from both patients and controls to obtain serum for serologic studies of viral antibody levels, and other tests that may be indicated as data from the questionnaires are obtained.

Major findings: To date, 18 patients and controls have been interviewed, and the questionnaire has proved manageable.

Significance to biomedical research and the program of the Institute: This disease has been considered possibly infectious or immunologic. The observation of virus-like particles in patients lends support to this

assertion. If the disease is an acquired infection, a case-control study may develop leads as to causation or predisposing factors.

Proposed course: A total of 50 cases and controls will be interviewed. Any suggestive data will be incorporated into another questionnaire and tested in a different population of cases.

Honors and Awards: None

Publications: None

Serial No. NDS (CF) - 73 E 2046
1. Collaborative & Field Research
2. Epidemiology Branch
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Clinical and Genetic Study of the Progressive Myoclonic Epilepsies

Previous Serial Number: None

Principal Investigator: Robert Stern, M.D.

Other Investigators: Roswell Eldridge, M.D.

Cooperating Unit: Department of Neurology, University of Pennsylvania School of Medicine

Man Years:

Total:	5/16
Professional:	1/4
Other:	1/16

Project Description:

Objectives: To study the clinical and genetic aspects of progressive myoclonus epilepsies and thereby establish a classification of these disorders leading to biochemical evaluation of specific disease entities.

Methods employed: Through neurology training programs and electroencephalographers in the United States, it was possible to ascertain thirty families with a diagnosis of progressive myoclonic epilepsy. Examination of affected individuals and their relatives, laboratory tests, and review of medical records provided a basis for a clinical and genetic classification of these disorders.

Major findings: On the basis of the examination of thirty-seven affected individuals in nineteen families the following classifications of these disorders were made:

Type IA - (Unverricht): Childhood onset, rapid progression, autosomal recessive inheritance, Lafora bodies often present.

Type IB - Childhood onset, slow progression, autosomal recessive inheritance, no Lafora bodies.

Type IIA - (Lundborg): Late adolescent onset, autosomal recessive inheritance, moderate or slow progression, occasional Lafora bodies.

Type IIB - Late adolescent onset, moderate to slow progression, autosomal dominant inheritance, neural hearing loss.

Significance to biomedical research and program of the Institute: Distinction between the various disease producing progressive myoclonic epilepsy and dementia is a pre-requisite for determing the specific metabolic defect in each disease. In a practical sense this classification should be of interest to clinicians faced with diagnosis of disorders including myoclonus and epilepsy. In addition, such a study provides a model for the genetic study of seizure disorders. Documentation of inheritance pattern in these disorders permits improved genetic counseling for members of these families.

Proposed course: This section, through its identification of affected individuals, is able to aid interested investigators in obtaining materials for study. We have aided biochemical studies now underway at the University of Pennsylvania. Increased biochemical knowledge of any one of these disorders should improve understanding of mechanisms of epilepsy and degenerative CNS disorders.

Honors and Awards: None

Publications: Stern, R., Eldridge, R.: Clinical and Genetic Study of the Progressive Myoclonic Epilepsies. (To be presented to the American Academy of Neurology Annual Meeting, April 1973).

ANNUAL REPORT
July 1, 1972 through June 30, 1973
Infectious Diseases Branch, C&FR
National Institute of Neurological
Diseases and Stroke
John L. Sever, M.D., Ph.D.

Introduction

The goal of the Infectious Diseases Branch is to carry out planned, directed research programs concerned with infections which damage the human nervous system. The Branch was recently divided into three sections: 1) Immunochemistry and Clinical Investigations, 2) Virology and Bacteriology, and 3) Experimental Pathology. These sections utilize the techniques of immunology, clinical investigations including human volunteers and clinical trials; virology, bacteriology, mycoplasmaology, neurovirology and electron microscopy; as well as experimental pathology with non-human primates.

The program segments are: a) Perinatal, b) Acute and c) Chronic. In each segment we are concerned with 1) etiology and diagnosis, 2) treatment, and 3) prevention.

The research activities in the program segments include:

1. Perinatal

Develop and utilize large scale microbiological methods to study the relation between viral, bacterial, mycoplasma and protozoa infections in the perinatal period and birth defects, related abnormalities and pediatric neurological diseases. Investigate approaches to early diagnosis, treatment and prevention using combined laboratory and clinical studies.

2. Acute

Investigate agents which may be responsible for acute neurological diseases such as meningitis, encephalitis, Bells' Palsy, and tic douloureux as well as possible methods for rapid diagnosis, treatment and prevention.

3. Chronic

Study chronic neurological diseases such as multiple sclerosis, amyotrophic lateral sclerosis and subacute sclerosing panencephalitis using combined tissue culture, immunological, serological, electron microscopic and clinical approaches for possible infectious etiologies. Whenever possible explore methods for early diagnosis, treatment and prevention.

A. Findings

1. Perinatal

a. Gammaglobulin as Prophylaxis Against Rubella Induced Congenital Anomalies

The use of gammaglobulin with various titers of antibody to rubella demonstrated in Hawaii that clinical rubella could be prevented in pregnant women exposed to this disease by the early use of high titered gammaglobulin. It appeared that congenital rubella was also reduced since none of the women who received high titered gammaglobulin had children with this disease. Because of the small number of patients, however, further studies of this type will be necessary to document the effectiveness of high titer rubella gammaglobulin for the prevention of congenital infection and damage.

b. Toxoplasmosis In Women From Puerto Rico and Their Children

High rates of antibody to toxoplasmosis among pregnant women from Puerto Rico living in New York City were observed in our investigations with patients from the Collaborative Perinatal Research Study. Increased rates of fetal disease due to congenital toxoplasmosis were also found in this population. Our further studies in Puerto Rico showed high rates of infection in children and adults. Children became infected primarily between six months and three years of life. These infections were not associated with parasitic diseases. The mode of transmission was not established for these populations but cats were postulated to be a likely source of infection. Raw meat as a source of infection could be excluded since these individuals rarely have meat of any type in the diet.

c. Hepatitis and Mongolism

Several reports have postulated that mongolism may be related, at least in part, to hepatitis early in pregnancy. Studies of Australian antigen and antibody using specimens from the Collaborative Perinatal Research Study showed no evidence of association between serum hepatitis and mongolism. It remains possible, however, that infectious hepatitis may be involved in this disease since the methods available do not permit specific testing for this type of hepatitis.

d. New Tests for Cytomegalovirus, Herpes I and Herpes II

The available complement fixation tests for cytomegalovirus for Herpes I and Herpes II viruses have not been satisfactory since frequent cross reactions are encountered. This is particularly important in studies of congenital disease caused by these agents. We have been able to develop more specific tests using the indirect hemagglutination methods. The new methods have now replaced the complement fixation tests in our laboratories as well as many other laboratories throughout the world.

e. Safety of Rubella Vaccine During Pregnancy

Vaccines for rubella are designed to prevent congenital rubella. Unfortunately since live vaccines are used, there is a risk that the vaccine virus itself may transmit to the fetus and cause damage if given during pregnancy. Our studies of the Cendehill vaccine virus showed no evidence of transmission to the fetuses studied and only rarely was the virus found in the placentas. This gives encouragement to the further studies on the possible use of the vaccine in women of child bearing age.

f. Mycoplasma Infections and Fetal Disease

Several reports indicate that mycoplasma infection during pregnancy may result in fetal infection, disease and prematurity. In addition, infection of the ovaries has been postulated as a cause of genetic damage to the ova. Our studies show high rates of fetal-maternal infections. Fetuses obtained at abortion were also infected but amniotic fluid was sterile. These investigations indicate that as with Herpes virus infections of the newborn, mycoplasma infections are usually acquired at birth and attention must be focused at longitudal observations of the infected children. Other studies of uterine and ovarian specimens are also needed to define the frequency of infection of these tissues and the possible association with prior abnormal pregnancy outcomes.

g. Experimental Production of Hydrocephalus with Influenza-A Virus in Monkeys

A number of studies have suggested that Flu-A virus may cause fetal infection, malformations and respiratory distress in the newborn. The data, however, has not been clear since the infection of the mother was usually based on clinical findings alone.

Experimental studies with pregnant rhesus monkeys in our laboratory now clearly demonstrate the production of hydrocephalus in the fetus by inoculation with flu-A virus at 100 days gestation. These studies are being extended to other viruses and extensive investigations of human clinical material from the Collaborative Perinatal Research Study.

2. Acute

a. Spinal Fluid Lactic Dehydrogenase (LDH) for Rapid Diagnosis of Meningitis

The determination of LDH levels in the spinal fluid is of considerable value in the rapid diagnosis of meningitis. This is important since therapy must be instituted immediately. New "bedside" tests for this enzyme now appear to be possible and are being developed for routine use.

b. Treatment of Herpes Encephalitis and Congenital Herpes

Herpes encephalitis and congenital herpes are severe, frequently fatal infections of adults and infants. Treatment with drugs such as ARA-A and ARA-C appear to have some potential value. Our studies now indicate that ARA-A is less toxic and may be superior to ARA-C. Further investigations in sub-human primates are being initiated at this time.

3. Chronic

a. Subacute Sclerosing Panencephalitis (SSPE) Registry

Joint studies with Dr. Jabbour at the University of Tennessee indicate that over 350 patients with SSPE during 1960-1963 have now been identified. More than 30 received measles vaccines. The mean interval between measles and SSPE is six years. The average patient lives about one year after diagnosis. There have been no spontaneous cures. We are particularly interested in the effect of mass measles immunization campaigns begun in the late 1960's. We hope there will be a decrease in the frequency of SSPE in the near future as a result of these vaccine efforts. The establishment of the registry should provide us with the longitudinal information neccessary to determine the possible effectiveness of the vaccine in the prevention of SSPE.

b. Chronic Cytomegalovirus Infections

Pregnant women, children and individuals with cancer have been found to be infected with cytomegalovirus for long periods of time. At least 3-8% of pregnant women have the virus present in the urine and vagina at the time of delivery. We have found that about 1-3% of newborns are infected. Children frequently become infected and excrete the virus for many months. Individuals with leukemia were found to have high rates of infection and this may have contributed to some of the clinical findings and disease in these patients.

c. Multiple Sclerosis and Infection

Recent reports have suggested that myxovirus-like viruses may be closely associated with multiple sclerosis. In our studies we have found by electron microscopy particles that resembled paramyxoviruses in the brain of an MS patient. We are involved in attempts at identifying the particles by the use of peroxidase labeled antibody for electron microscopy and direct virus isolation. Three brain specimens from MS patients have been studied for virus isolation and no virus was recovered using routine techniques. The new methods of cocultivation and IUDR shock are now being employed in the further study of MS tissues.

d. Treatment of Subacute Sclerosing Panencephalitis

Attempts to treat patients with SSPE using transfer factor were not successful. All other approaches to date have also been of no value. The use of BCG to stimulate cellular immunity and plasmapheresis for the possible "wash out" of cellular inhibitors to cellular immunity is being considered in clinical protocols at this time.

e. New Methods for Detecting Specific Cellular Immunity to Virus Diseases

Joint research between investigators in the Infectious Diseases Branch and Georgetown University Medical Center have lead to highly specific and sensitive new tests for cellular immunity to virus infections such as measles and German measles. These tests are extremely valuable in our studies of immune deficiencies in patients with chronic infections of the nervous system. New findings indicate suppressed cellular immunity in rubella and subacute sclerosing panencephalitis. Further studies of congenital herpes and multiple sclerosis are in progress.

B. Studies in Progress

1. Perinatal Infections

a. Collaborative Study

(1) The Phase II Studies

Particular emphasis is being placed on three studies through FY 1975. Reports on this work will be given at the IV International Congress on Malformations in Vienna in September 1973. The final report will be available in 1975. The studies include three approaches to the Collaborative Study data on 58,000 pregnancies.

First- Clinically Recognized Infections, Serologically Confirmed When Possible and Pregnancy Outcomes.

There were approximately 5,000 viral, bacterial, protozoa and parasitic and fungel infections in the study population. These data are being correlated with clinical data for the children.

Second- Serological Studies Using Eleven Antigens for 4,000 Pregnancies with Abnormal Outcomes and 4,000 Matched Controls.

Paired sera are being tested under code. Tests are approximately 50% complete.

Third- High IgM Cord Levels and Abnormal Babies.

A total of 32,000 cord sera have been tested. We have found 2,200 with high IgM. These sera are now being studied for specific antibody to syphilis, toxoplasmosis, rubella, cytomegalovirus and herpes type II.

(2) The Phase III Studies

Special Studies of Infection, Malformations

Studies of neurological damage relating to sepsis and toxoplasmosis are in progress. In addition, a small study of potato ingestion and birth defects are being conducted. Further serological studies will be initiated in depth when Phase II is completed.

b. Clinical Investigations

(1) High Risk Babies - University of Tennessee

Admissions to the high risk nursery at the University of Tennessee are being studied in detail. IgM levels are determined serially on all babies. Sick and damaged infants are tested for evidence of infection.

(2) Infertility, Abortion and Infection - UCLA, Kaiser

Women with infertility or repeat abortions are being tested for evidence of infection. Sets of specimens are studied along with sera for virus isolation and antibody. Previous longitudinal studies of infections are being analyzed at present.

(3) Mycoplasma Infections and Abortions, Prematurity and Malformations

Direct isolation of mycoplasma has been found to be much more sensitive and specific than serological approaches. Mycoplasmas have been associated with abortions, prematurity, and probably with malformations. Our study at the Naval Medical Center is directed at the role of mycoplasma and cytomegalovirus in abortions, prematures and malformations. Isolations are being made from the placenta, fetal tissue and endometrium. Specimens of ovary taken at hysterectomy are now becoming available for study.

c. Experimental Animals

(1) Malformations In Virus Infected Monkeys

Studies of hydrocephalus and cataracts produced by Flu-A and Venezulian Equine Encephalitis viruses are being extended to related viruses and vaccine viruses. Time and route of inoculation is being varied. These studies are particularly rewarding and provide tools for the investigation of pathogenesis of these diseases.

(2) Immunosuppression and Infection

Joint studies supported by the National Cancer Institute permit the investigation of immunosuppression and fetal infections in pregnant monkeys. Immunosuppression agents increased the likelihood of congenital infection and permit investigations of a variety of agents.

(3) Potatoes and Birth Defects

Pilot studies on possible teratogenic effects of potatoes have been initiated in pigs.

(4) Immune Responses in Fetal Monkeys

The development of antibody, cellular resistance, and interferon is being studied in fetal monkeys which are infected in utero. This provides for the first time direct information on immune responses for congenital infections in primates.

d. New Methods

New tests for viral antibody using radioimmune methods are being developed for our studies. Rapid methods for IgM and specific IgM are being investigated and perfected for clinical use.

2. Acute Infections

a. Spinal Fluid LDH For Rapid Diagnosis of Meningitis

New, rapid methods for CSF levels of the enzyme LDH are being developed to permit the more rapid diagnosis of bacterial meningitis.

b. Treatment For Herpes I, II

Drugs such as ARA-A, ARA-C, and 5-IDU are being investigated for treatment of congenital and acquired herpes I and II infections. We are using owl monkeys in the studies of these drugs since this monkey is particularly susceptible to herpes I and II.

3. Chronic Infections

a. Clinical Studies

(1) SSPE Registry

We are continuing a joint effort with Dr. Jabbour at the University of Tennessee to determine the number of patients each year reported

with SSPE. One hundred medical centers are contacted yearly. We are interested in possible changes in the epidemiology of SSPE with use of vaccines for rubeola.

(2) Treatment of SSPE

The presence of a serum inhibitor of cellular response to measles in patients with SSPE is being studied in detail. If this inhibitor can be removed we will institute clinical studies on the possible benefits of this approach in man.

b. Clinical - Laboratory Studies

(1) New Methods For Specific Viral - Cellular Immunity

Methods using Chromium 51 release for infected cultures for determining cellular immunity have been developed for several viruses. We are extending this to other infections and clinical studies of immune responses to chronic infections and neurological diseases including MS, ALS, and SSPE.

(2) Bell's Palsy and Trigimal Neuralgia

Attempts are being made to isolate infectious agents which may be responsible for Bell's Palsy and Trigimal Neuralgia. Both virus isolati and serological approaches are being used.

(3) MS, ALS, AND Guillain-Barré - Serological Studies

Serological surveys for a variety of infectious agents are being conducted for these three diseases. Direct and indirect fluorescence studies are being conducted for MS and ALS using fresh tissues.

(4) Electron Microscopy - MS AND SSPE

Direct electron microscopy studies as well as specific viral electron microscopy studies using peroxidase labeled antibody are being conducted with MS and SSPE tissues.

(5) Virus Isolation - MS and ALS Tissues

Tissues from patients with MS and ALS are being studied for the presence of virus using cocultivation, IUDR shock, and serial passage. Tests for viruses in the cultures include electron microscopy, fluorescence, inoculation of animals for antibody, hemadsorption, and observation for cytopathic effect.

(6) Experimental SSPE - Immune Reponse

Hamsters are being inoculated with SSPE virus to produce SSPE like disease. These animals are "treated" with immune serum or cells to determine the effect of these immune responses on the production of the disease.

C. Conclusion

The research activities of the Infectious Diseases Branch are divided into three areas: Perinatal, Acute, and Chronic. The perinatal clinical research is further divided into 1) studies relating to the Collaborative Perinatal Research Study and 2) cooperative and other studies. The total perinatal area accounts for more than 65% of the research effort while acute infections involve about 15% of the effort and chronic infections 20%.

Major reports are being developed on Phase II of the Collaborative Perinatal Research Study. New experimental findings with infections in monkeys are providing tools for the study of infections and malformations. Acute infections are being studied with new methods for diagnosis and chemotherapy. Chronic diseases of the central nervous system are being investigated with newly developed tests for specific cellular resistance, electron microscopy and brain tissue cultures.

CONTRACT NARRATIVE
Infectious Diseases Branch, C&FR, NINDS
Fiscal Year 1973

DEPARTMENT OF PEDIATRICS, UNIVERSITY OF TENNESSEE (PH43-68-17)

Title: Identification of Specific Antibodies in the Serum IgM Fraction of High Risk Infants

Contractor's Project Director: Dr. Sheldon Korones, M.D.

Current Annual Level: $25,720.

Objective: The study is designed to assess the validity and clinical usefulness of specific tests for serum IgM for the identification of antibodies to rubella, cytomegalovirus, toxoplasmosis, herpesvirus and syphilis. When these diseases are identified as being present in the children, appropriate therapeutic approaches are used.

Major Findings: The study was initiated during the last fiscal year. Approximately 1,000 infants have been tested to date. Serial blood specimens are collected twice each week from each study infant for the duration of the nursery stay. Venous blood is obtained from infants with IgM concentration is in excess of 20 mgm per cent in the first two weeks of life and 30 mgm per cent between the third and fourth weeks. Virus cultures are obtained from all infants with high IgM levels. Clinical evaluation data is being maintained for correlation with the serologic findings.

Significance to NINDS Program and Biomedical Research: The detection of high risk infants with congenital infections is hampered by the lack of specific tests to identify these children. Recent studies have indicated that elevated IgM levels are present in at least 80 per cent of children with congenital infection. The present studies will provide direct information as to the usefulness of specific IgM antibody determinations for five major infectious agents as a means of detecting not only the possible presence of congenital infection but the specific infection which requires treatment.

Proposed Course of Project: The project is completing its first year. We anticipate at least two additional years of study during which a total of 2500 infants will be examined.

CONTRACT NARRATIVE
Infectious Diseases Branch, C&FR, NINDS
Fiscal Year 1973

THE JOHNS HOPKINS UNIVERSITY (PH43-68-710)

Title: Longitudinal Surveillance of Children with Congenital Infection

Contractor's Project Director: Dr. Janet Hardy, M.D.

Current Annual Level: 0

Objectives: The contractor will maintain longitudinal surveillance of children born during the 1964 epidemic of rubella who have been identified as having congenital rubella. These children were originally studied in conjunction with the Infectious Diseases Branch of the National Institutes of Health in 1964-1966. Serial specimens are being obtained from the children for virus isolation and antibody determinations. Clinical evaluation is being obtained repeatedly so as to determine the total clinical findings for these children throughout a twelve-year period of observation.

Major Findings: During the first four years of observation, it became obvious that many children who were initially thought to be normal showed damage after the passage of time. In addition, high antibody levels found in the first year of life frequently decreased to low or undetectable levels, thus making the retrospective diagnosis of the disease impossible. The present findings indicate much higher rates of damage than had been anticipated. Most of this damage is related to the more subtle manifestations of mental motor retardation and mild to moderate hearing deficits.

Significance to the NINDS Program and Biomedical Research: Congenital rubella infection is a preventable disease. The wide spread use of rubella vaccines should prevent the occurrence of epidemics and thus greatly reduce the frequency of the disease. Damage due to the infection however , has only been documented in detail in recent years. Severe congenital infection has been noted since 1941 to be associated with severe manifestations of the disease. More subtle disease entities however, are now being recognized and it is evident that many of the children heretofore classified as "etiology unknown" are the result of unrecognized intrauterine infection with rubella.

Proposed Course of the Project: The project will continue through the twelve years of observation with a repeated clinical and laboratory findings for final presentation in 1976.

CONTRACT NARRATIVE
Infectious Diseases Branch, C&FR, NINDS
Fiscal Year 1973

THE UNIVERSITY OF CALIFORNIA, LOS ANGELES (NIH-NINDS-69-4)

Title: Infections in Pregnancy and Infertility

Contractor's Project Director: Dr. Margaret Jones, M. D.

Current Annual Level of Support: $29,138.

Objectives: The contractor will analyze clinical and serological data collected during the prospective study of the effect of maternal viral infections and pregnancy outcome in approximately 4500 women. Serial serological data is available for five viruses for this population. This study was conducted in 1959-1963 and there is now an opportunity for long term followup for the children whose mothers had serological evidence of infection.

The contractor will also determine the frequency of viral infections and chronic intrauterine infections among cases of infertility and controls as a study of the causes of infertility.

Major Findings: Data has been tabulated for patients with seroconversions and four fold increases in antibody to influenza, cytomegalovirus, rubella, mumps, and herpes. This is being analyzed in relation to time in pregnancy and pregnancy outcome. Matched controls are being used in this analysis. Multiply infected women are being analyzed separately. In addition, the prevalence of antibody is being determined for each season and the persistence of antibody is being evaluated.

Specimens from women who are being seen because of infertility are under test. A total of 50 women have been studied to date. Blood specimens, throat swabs, cervical swabs and endometrial biopsies are being used.

Significance to NINDS Program and Biomedical Research: Congenital infection is a preventable cause of severe central nervous system and other diseases in children. Elimination of this risk factor is an exceedingly worthwhile approach to the prevention of damage to children and the related social and economic problems resulting from this type of damage. Infertility is often related to viral and chronic intrauterine infection and is directly associated with the production of damaged children. These studies, therefore, will provide us opportunities for determining which agents are the most frequent causes of infertility and fetal damage.

Proposed Course of Project: The project will run for two more years. The emphasis will continue on analysis of the clinical data for the 4500 women. We will include cord IgM data during the next year.

Specimens from women with infertility will be studied in detail. An additional 50 women will be included during the next year.

Serial No. NDS (CF)-57 ID 402
1. Infectious Diseases Branch
2. Section on Immunochemistry and Clinical Investigations
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Immunology of Perinatal Infections

Previous Serial Number: Same

Principal Investigators: Dr. John L. Sever, IDB, NINDS
Dr. David A. Fuccillo, IDB, NINDS
Mrs. Anita Ley, IDB, NINDS
Mrs. Renee Traub, IDB, NINDS
Mrs. Mary Ruth Gilkeson, IDB, NINDS
Mrs. Flora Moder, IDB, NINDS
Mr. David Rubinstein, OB, NINDS
Dr. Jonas Ellenberg, OB, NINDS

Other Investigators: Dr. Gabriel Castellano, MBA
Dr. Janet Hardy, Baltimore, Md.
Dr. Margaret Jones, Los Angeles, California
Dr. Paul Terasaki, UCLA, Los Angeles, California

Cooperating Units: Collaborating Institutions in the Perinatal Research Study
Laboratory of Infectious Diseases, NIAID
NICHD

UCLA Medical School
Johns Hopkins University Medical School
Kaiser Hospital, Los Angeles

Man Years:

Total:	13.5
Professional:	5.5
Other:	8

Project Description:

Objectives: The purposes of this study is to determine insofar as possible the role of infections and immunity in the production of abnormal pregnancy outcomes. To accomplish this, 12 collaborating institutions in the Perinatal Research Study plus two special cooperating groups in separate studies have been obtaining specimens of blood and tissue throughout pregnancy, at delivery, post partum, and at set intervals thereafter. These sera are tested to determine the antibody responses of the patients during pregnancy and post

partum and then to relate this serological information to the clinical data for the pregnancy and child. In addition, serum specimens from the children were obtained at one year of age from 10,000 study pregnancies. Sera, throat swabs, and urine specimens were also obtained from approximately 5,000 pregnancies. Placental specimens were obtained from 2,500 pregnancies. In special cases when congenitalinfection is suspected on the basis of clinical or laboratory findings, throat swabs and blood specimens are obtained from the children. Immunoglobulin determinations are performed with the cord blood specimens from the children and specific antibody determinations are also made with these specimens.

Methods Employed: To accomplish this program, blood specimens were obtained from pregnant women at set intervals throughout pregnancy and post partum. Completeness of the sets of sera is determined at the Serum Center of the Infectious Diseases Branch. Data for the 58,000 patients in the Collaborative Perinatal Research Study show that specimens are available from 94.2% of the patients. An average of 5 blood specimens is available for each patient. Each specimen consists of 4 vials with 3 ml of serum in each. For this study then, there are approximately 300,000 serum specimens and almost a million and a half vials of sera. There are an additional 5,000 patients studied to date at the Kaiser Hospital in Los Angeles and approximately 3,000 under study at the Johns Hopkins Medical School in Baltimore, Maryland. All specimens are stored at -20°C until tested and complete filing record concerning basic patient information and the status of the serum available is maintained through a computer system by the Serum Center of the Branch.

In addition to the serum specimens, serial urine and throat specimens were also obtained on a large majority of the patients in the two special studies. These are being studied for direct virus isolation along with swabs obtained from the children at the time of birth.

To date, approximately 50 publications have resulted from the analysis of the data from these studies. The serological method most frequently employed is the complement fixation test with the use of viral antigens. The test is very versatile and can be performed rapidly and provides broad coverage for a great many of the more than 130 viruses which are known to be of importance to man. Antigens were prepared for most of these viruses and tests of specificity were conducted with animal sera. In addition to the complement fixation method, hemagglutination inhibition tests are used for many viral serological determinations when greater specificity is needed, neutralization methods are employed. Indirect fluorescence is also used for some of the studies. Virus isolation is conducted with tissue culture or inoculation of experimental animals.

All tests are reproduced completely and a minimum of 95% agreement within two-fold variation as required. All sera showing significant changes in antibody, together with any sera which did not reproduce are tested the third time.

At present, we are engaged in a large scale two-year study designed to investigate infection and immunity in relation to 4,000 normal children in the study and 4,000 matched controls. The crude print-out of abnormal patients has been

obtained from the Collaborative Perinatal Research Study (PRB) and this is being reviewed in detail by nurses and physicians from the IDB for more complete information.

From study records, the specific abnormalities under study include abortions, stillbirths, cataracts, congenital heart disease, neonatal deaths, low birthweight (1,000-1,500, 1,500-2,000 grams), IQ below 50, IQ 50-69, enlarged liver, malformations, retarded gross motor development, retarded fine motor development, hearing deficit in both ears, visual impairment, cranial or peripheral nerve damage, cerebral palsy, delayed motor development, hypotonia with poor deep tendon reflexes, non-febrile seizures, dyskinesia and ataxia, hearing deficit in one ear, and elevated bilirubin. The specimens from the mothers of these children and from the children themselves along with carefully matched controls are being studied for antibody to 11 antigens. These antigens include influenza A, rubeola, rubella, mumps, Coxsackie B_3, Coxsackie B_4 varicella, toxoplasmosis, cytomegalovirus, herpes Type I, and herpes Type II. All of these agents are known or suspected to be responsible for damage in the perinatal period. All laboratory work is being performed under code. The data is being analyzed by Mrs. Gilkeson and Mr. David Rubinstein.

A second approach is the study of reported viral, bacterial, and protozoal infections in pregnant women in the study. Serological tests are used to document these reports. The data is then correlated with the pediatric findings. Approximately 2,500 cases of reported viral infections, 3,000 bacterial infections and several hundred protozoal infections, are under investigation. Clinical data is being abstracted, serological tests are being performed in order to document these infections. There are also approximately 1,200 patients identified with a positive serology for syphilis. These are being studied in detail.

The third approach is to identify the children with elevated IgM levels in the newborn period and then to correlate these findings with pregnancy outcome, clinical performance of the child, and specific serological tests for IgM antibody. Almost 32,000 cord sera have been tested for IgM antibody and approximately 2,000 show elevated levels. These are now being studied in detail.

Major Findings: Placentas from approximately 2,500 births collected at five collaborating institutions after the 1964-65 rubella epidemic were tested for presence for rubella virus. The virus was isolated from 13 (0.52%) of the specimens. The study then permitted the identification of the rate of congenital infection to be approximately 1 in 200 pregnancies. The children were studied in detail and evidence for congenital rubella was documented at birth and at four months of age. Rubella virus was isolated from many of these children. This approach permitted the prospective investigation of the frequency of congenital infection and associated clinical findings.

A serologic survey for Australia antigen and antibody was conducted to determine the frequency with which this hepatitis-associated antigen was present in pregnant women with clinical hepatitis and the frequency with which the infection was transmitted to the developing fetus. Eighteen women

were identified who had had hepatitis during pregnancy. Two of these women had the antigen present in the serum specimens which were submitted for study. One of the children had antigen in the cord serum, but was without clinical hepatitis. These studies demonstrated for the first time the vertical transmission of the Australia antigen associated with hepatitis. None of 27 serum specimens from pregnant women who delivered children with Down's syndrome was positive for the antigen. This finding was important because of the previously reported possible association of hepatitis with the occurrence of Down's syndrome. In a further investigation of 262 institutionalized patients with mental retardation of various etiologies, 10% were positive to Australia antigen by the complement fixation test, while 34.1% of 138 patients with Down's syndrome were positive. The association of Australia antigen with Down's syndrome then appears to be an acquired phenomenon not related to maternal infection and not present in the newborn.

A seroepidemiologic study of toxoplasmosis was conducted in the Collaborative Perinatal Research investigation in which we found a high prevalence of antibody to toxoplasmosis among women from Puerto Rico. Studies conducted by our laboratory with Dr. Dolores Mendez-Cashion of the University of Puerto Rico School of Medicine indicated that children become infected with toxoplasmosis in Puerto Rico between six months and two years of age. Approximately 50% of the dogs and cats studied had evidence of infection. In this study of antibody and intestinal parasites in Puerto Rican children, it was found that a third of the children had evidence of parasitic infestation by two years of age. Fifty-one percent of the mothers and 8% of the children were found to have antibody to toxoplasma. Attempts to isolate toxoplasma in mice by feeding material from children with stools was unsuccessful and there was no correlation between contact with animals and presence of antibody to toxoplasmosis. The further longitudinal study of children ages 12-36 months coupled with study of their animal contacts is now in progress.

Cytotoxic (HLA) antibodies present in the serum of women with various abnormal pregnancy outcomes and matched controls were studies with Dr. Paul Terasaki, UCLA. A total of 3.5% of women who were pregnant for the first time had antibodies, however, this increased to 16 to 21 percent when the gravidities were three or more. These studies indicate that antifetal tissue antibodies are present in these pregnancies.

Collaborating Studies: A primary deficit in the data from the Collaborative Perinatal Research Study has long been recognized as the late registration of the study's patients. Since only 20% of the patients registered during the first trimester of pregnancy, it is impossible to document adequately the infectious disease experience of the patient's during the first trimester. To provide data on the first trimester of pregnancy and high-risk pregnancies, an additional cooperative study was initiated with the Infectious Diseases Branch. This is a study of viral infections in pregnancy of Dr. Margaret Jones UCLA, at the Kaiser Hospital, Los Angeles, California.

A second study was initiated at the Kaiser Hospital, Los Angeles, California to study infection in relation to infertility. Approximately 50 women

are being studied each year. In each case sera, throat swabs, cervical swabs and endometrial biopsy specimens are obtained and sent to the IDB laboratories for detailed study.

A second deficit of the Perinatal Research Study has been the lack of serial blood and tissue specimens from children with known congenital infection. For this reason, a special investigation was initiated with Dr. Janet Hardy at the Johns Hopkins Medical School to provide longitudinal surveilance of children with congenital infections to determine the clinical course, as well as the antibody levels, immunologic responses, and shedding of infectious agents in these children.

Significance of the Program to the Institute: The use of micro-serological techniques for a large group of new viruses provides an opportunity to investigate the disease caused by viruses which are either difficult to isolate or resistant to evaluation because the clinical effects are delayed until a long time after the infection has subsided. In addition, the availability of new immunologic techniques provides the unique opportunity to detect immunologic deficits and to determine the presence of intrauterine infections on the basis of immunologic response. This data can then be correlated and analyzed as in relation to the possible causes of birth defects. The application of this type of analysis has provided valuable information on the epidemiology of virus infections in relation to abnormal pregnancy outcomes and is constantly giving us new insights into the causes of damage to the central nervous system and possible means of prevention of this and other damage to the developing fetus and newborn.

Proposed Course of the Project: The combined immunologic virologic program will continue at a very intense rate during the next two years. During that time we plan to perform the great majority of tests which are needed in relation to the Collaborative Perinatal Research Study. Subsequent testing will then be limited to special studies in depth and the intensive investigations in relation to the cooperative study at the Kaiser Hospital in Los Angeles and at the Johns Hopkins Medical School.

The three approaches which are being emphasized now include:

1. A special two-year commitment to perform serological tests on 4,000 abnormal pregnancies and 4,000 matched controls using 11 antigens. The abnormal children have been identified and the laboratory is now approximately half way through the testing.

2. Special testing of IgM levels from 32,000 cord sera from children in the Collaborative Study and in the cooperative studies. This work provides an index for identifying children with possible congenital infections so that more specific testing can then proceed. These investigations are now well under way and will be continued during the next two years. Approximately 2000 cord sera have high IgM levels. These are being tested for specific antibody to rubella, toxoplasma, cytomegalovirus, herpes II and syphilis.

3. A correlation of clinically reported infections in pregnancy with serological findings for the pregnancy, immunologic determinations, and pregnancy outcome. These studies are now in progress and should be completed for the most part within the next year.

New rapid methods for determining IgM levels and specific IgM antibody will be studied.

Honors and Awards: None

Publications:

McCallin, P. F., FACOG, Fuccillo, D. A., Ley, A. C., Gilkeson, M. R., Traub, R. G., and Sever, J. L.: Gammaglobulin as Prophylaxis Against Rubella-Induced Congenital Anomalies. Ob. & Gyn., Vol. 39, No. 2, February 1972, pp. 185-189.

Newman, S. J., Fuccillo, D. A., Sever, J. L., London, W. T., and Mendez-Cashion, D.: A serological and Epidemiological Study of Toxoplasmosis in Puerto Rican Children. Clinical Proceedings of Children's Hospital, Vol. 28, no. 2, February 1972. Pp. 47-53.

Sever, John L.: The Expectant Mother: Exposure to Infectious Diseases. Pediatric Portfolio, Vol. II. No. 3, 1972.

Serial No. NDS (CF)-61 ID 835
1. Infectious Diseases Branch
2. Section on Immunochemistry and Clinical Investigations
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Clinical Investigations with Human Volunteers and Other Populations Using Viruses, Vaccines, and Chemotherapeutic Agents.

Previous Serial Number: Same

Principal Investigators: Dr. John L. Sever, IDB, NINDS
Dr. Jerome Kurent, IDB, NINDS

Other Investigators: Dr. David A. Fuccillo, IDB, NINDS
Dr. Michael Blaese, NCI
Dr. Robert Miller, NIH, NCI

Dr. Paul McCallin
Kaiser Hospital, Hawaii

Cooperating Units: Bureau of Prisons, Dept. of Justice
Petersburg Federal Reformatory
(Dr. J. S. Goldberg, Chief Medical Officer)

Man Years:

Total	1
Professional:	.5
Other:	.5

Project Description

Objectives: To study new prophylactic and therapeutic materials for the prevention and control of infectious diseases. To study the safety, antigenicity, communicability, and immunogenicity of candidate vaccines.

Methods Employed: Human volunteer studies are conducted in collaboration with the Federal Bureau of Prisons. These studies are reviewed and approved by the Clinical Research Committee and the Medical Board of the National Institutes of Health and the Medical Review Board of the Federal Bureau of Prisons.

Vaccines and chemotherapeutic agents which appear to warrant further investigations are then utilized in clinical studies of other special patient groups. In addition, clinical studies of epidemic are investigated as a means of identifying the role of infectious agents in the production of perinatal and pediatric disease states.

Major Findings: The drug cytosine arabinoside was studied in two infants with congenital cytomegalovirus infection. One case appears to have had some modification of virus excretion during the period of therapy. The second patient stopped excreting cytomegalovirus just prior to therapy, but continued to harbor the virus in the white blood cells after therapy. Toxicities noted included vomiting, neutropenia, and thrombocytopenia.

A total of 7 patients with SSPE were treated with transfer factor under our protocols at the Clinical Center. There was evidence of transfer of delayed hypersensitivity but no clinical response.

A major institutional epidemic of infectious hepatitis was studied at the Lynchburg Training School, Lynchburg, Virginia. Chronic carriers of Australia antigen were identified and found to be susceptible to infectious hepatitis. This observation provided additional evidence that serum hepatitis, which is not associated with the antigen, are immunologically dissimilar and caused by different etiologic agents. The institutional frequency and pattern of Australia antigenemia was reported. The antigen was present in 23 percent of patients with Down's syndrome and in 6 percent of other mentally retarded patients. The antigen was more prevalent in younger patients than in older patients and twice as frequent in males as in females.

Significance of the Program to the Institute: Volunteers studied provide the basic data necessary to evaluate potential chemotherapeutic agents and vaccines. They also provide invaluable information on the course of infection in man. Utilization of this information then in clinical studies provides the necessary bridge to the implementation of therapeutic approaches which originated in the laboratory, are brought through the volunteer studies and then finally are taken to patient population.

Proposed Course of the Project: The additional studies will be performed utilizing rubeola vaccines to determine antibody response. Rubeola antibody in SSPE patients will be studied in detail. Clinical studies will continue on utilization of various drugs for the prevention of treatment of perinatal, acute and chronic infections in man. The possible use of plasmapheresis to "wash out" inhibitor in serum of patients with SSPE will be considered. In addition, the use of new methods for determining spinal fluid lactic dehydrogenase levels in CSF will be studied in relation to the early diagnosis of meningitis.

Publications:

Kraybill, E. N., Sever, J. L., Avery, G. B., and Movassaghi, N.: Experimental Use of Cytosine Arabinoside in Congenital Cytomegalovirus Infection. Pediatrics, Vol. 80, No. 3, pp. 485-587, March, 1972.

Serial No. NDS (CF)-61 ID 835

Dietzman, D. E., Matthew, E. B., Madden, D. L., Sever, J. L., Rostafinski, M., Bouton, S. M., and Nagler, B.: The Occurrence of Epidemic Infectious Hepatitis in Chronic Carriers of Australia Antigen Pediatrics, Vol. 80, No. 4, Part 1, pp. 577-582. April 1972.

Serial No. NDS (CF)-72 ID 972
1. Infectious Diseases Branch
2. Section on Experimental Pathology
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Experimental Animal, Tissue Culture, Histopathological and Serological Investigation of the Role of Viruses and other micro-organisms in the Perinatal Period

Previous Serial Number: Same

Principal Investigators: Dr. William T. London, IDB, NINDS
Dr. Amos E. Palmer, IDB, NINDS
Dr. John L. Sever, IDB, NINDS
Dr. David A. Fuccillo, IDB, NINDS

Other Investigators: Mrs. Anita C. Ley, IDB, NINDS
Mrs. Blanche Curfman, IDB, NINDS

Cooperating Units: Dr. Stephen Kent
Department of Pathology
George Washington University
Washington, D.C.

Dr. Neal H. Levitt
Virology Division
U.S. Army Medical Research Institute of Infectious Diseases
Fort Detrick, Frederick, Maryland 21701

Man Years:

Total:	3.9
Professional:	1.4
Other:	2.5

Project Description:

Objectives: To study the role of viruses and other microorganisms in the perinatal period, the infection of gravid and nongravid animals of several different species by parenteral routes with various viruses and other micro-organisms to determine the effects of these agents on the animals and their fetal tissues.

Attempt to recover inoculated agents from the various animals and fetal tissues and the correlation of these reisolation with time (in gestation) of inoculation and dosage given. Relate these findings with gross and histopathological findings. Correlate all of this information with serological findings.

Methods Employed: An investigation of the role of viruses and other microorganisms in the perinatal period by the continual use of experimental animals; tissue culture techniques; histopathological studies; and serological testing.

Special quarantine facilities are maintained to prevent feral animals going through quarantine from becoming infected with endemic diseases such as rubeola and cytomegalovirus. A serological defined breeding colony of Rhesus monkeys is maintained to provide susceptible pregnant females for inoculation.

Pregnant animals are being inoculated by various routes with viruses and other microorganisms, and held in isolation chambers throughout the experiment. These animals are being observed and serum samples, liver biopsy, spinal fluid and throat swabs are checked for evidence of disease and/or effects on fetal tissue.

Virus isolation investigations utilizing tissue culture to recover viruses from tissues and fluorescent antibody technique to study the location of virus infection produced in experimental animals are being employed.

Major Findings: The intracerebral inoculation into the fetus of time-pregnant rhesus monkeys with low passage influenza "A" Virus, produced infection with antibody response in both fetus and mother. Hydrocephalus was observed in 50% of 12 inoculated fetuses. Abnormalities were not seen in any of the control animals (4). The infection resulted in stenosis of either the aqueduct of Sylvius or the foramina of Monro. In two animals, both ducts were stenotic. An acute ependymitis with resulting stenosis of the above mentioned ducts is believed to be the cause of the hydrocephalus.

The vaccine strain (TC83) of Venezuelan Equine Encephalomelitis Virus produced hydrocephalus and congenital bilateral cataracts in 10 fetal rhesus monkeys (100%). The animals were inoculated intracerebrally at 100 days of gestation. No abnormalities were observed in 4 similarly inoculated control animals. Tissue from these animals are now being processed.

Significance to the Program of the Institute: A program using experimental animals, tissue culture techniques, and histopathological investigations compliments the strict serological approach being used on human sera obtained from the Collaborative Study and thus balances the investigations of the role of viruses and other microorganisms in the perinatal period. It presents the direct means of investigation of these agents which may contribute to perinatal pathology.

Proposed Course of the Project: Further studies using mumps Reo, vaccinia viruses inoculated into pregnant monkeys are in progress. Using the known teratogens Flu-A and VEE, additional pregnant animals will be inoculated. The time in gestation and route of inoculation will be varied to determine the optional time in gestation for teratogenic effects and if the virus can cross the placenta.

Honors and Awards: None

Publications: None

Serial No. NDS (CF)-65 ID 1270
1. Infectious Diseases Branch
2. Section on Immunochemistry and Clinical Investigations
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Toxoplasmosis: Serological and Clinical Studies

Previous Serial Number: Same

Principal Investigator: Dr. John L. Sever, IDB, NINDS

Other Investigators: Dr. Joseph S. Drage, PRB, NINDS

Cooperating Units: Section on Pediatric Neurology, PRB, NINDS

Man Years:

Total:	.4
Professional:	.2
Others:	.2

Project Description:

Objectives: This study relates to antibody titer in pregnant women to abnormal pregnancy outcomes. A total of 23,000 women have now been studied.

Methods Employed: The computer analysis of multiple variables relating to toxoplasmosis in pregnancy outcome has been used. The serologic data is being correlated with pregnancy outcomes.

Major Findings: Within the group of 47 patients with high titers or significant increases in antibody titers, 5 were found to have definite toxoplasmosis and 10 were suspected of having possible toxoplasmosis. There were increased rates of stillbirths and neonatal deaths. The acquisition of antibody to toxoplasmosis was strongly correlated with age and race.

Proposed Course of the Project: Publication of complete analysis will be persued this year.

Honors and Awards: None

Publications: None

Serial No. NDS (CF)-67 ID 1503
1. Infectious Diseases Branch
2. Section on Immunochemistry and Clinical Investigations
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Epidemiologic Studies of Perinatal Infections

Previous Serial Number: Same

Principal Investigators: Dr. John L. Sever, IDB, NINDS
Dr. Jerome Kurent
Mrs. Dorothy Edmonds, R. N., IDB, NINDS

Other Investigators: Dr. Jonas Ellenberg, OB, NINDS

Cooperating Units: Collaborative Perinatal Research Study
Kaiser Hospital, Los Angeles, California
Kaiser Hospital, Hawaii
Kaiser Hospital, Oakland, California

Man Years:

Total:	1
Professional:	1
Other:	0

Project Description:

Objectives: Utilize information from collaborative and cooperative studies to identify pregnancies complicated by maternal infections or infections in childhood. To further delineate these cases by serological testing and to determine the outcome of these pregnancies in relation to the infections which have been defined.

Methods Employed: Primary material utilized for these studies are the clinical records, serum and tissue specimens obtained during pregnancy and the pediatric period of observation. Serologic data developed by the immunology laboratories is correlated with the clinical information from the collaborating groups. Data is analyzed in cooperation with the Office of Biometry, NINDS.

Major Findings: Chronic infection has been demonstrated to be present in the central nervous system of children infected with rubella virus in utero. Possible importance of this in the production of permanent brain damage is now under investigation.

At least a dozen viruses have been identified as being of importance in the perinatal period. The specific rates of the occurrence of these infections appear to be excess of 5 percent of the total pregnancies. As many as 10 percent of the children with microcephaly and mental retardation may be associated with these perinatal infections.

Rubella vaccine has been studied in pregnant women. There has been no evidence of transmission of vaccine virus to the fetus when the Cendehill strain of virus was used.

Significance of the Program of the Institute: The identification of clinically reported cases of infection provides an immediate tool for analyzing the effects of perinatal and pediatric infections on the development of the child. The epidemiologic studies which are possible because of the large numbers of patients in the collaborative and cooperating studies provide the necessary data base for these types of investigations. Special studies are initiated in populations where high frequencies of infection or abnormal pregnancy outcomes have been noted.

Proposed Course of the Project: Special emphasis will be placed on analysis of data for neonatal meningitis and sepsis. There are approximately 50 cases of each with 5-8 year longitudinal followup observations which will be analyzed.

Honors and Awards: None

Publications:

Dietzman, D. E., Madden, D. L., Sever, J. L., Lander, J. J., and Purcell, R. H.: Lack of Relationship Between Down's Syndrome and Maternal Exposure to Australia Antigen. Amer. J. Dis. Child. 1972. Vol. 124, No. 2, Aug.

Serial No. NDS (CF)-69 ID 1729
1. Infectious Diseases Branch
2. Section on Virology and Bacteriology
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Role of Infections in Infertility, Abortions and Malformations

Previous Serial Number: Same

Principal Investigators: Dr. David A. Fuccillo

Other Investigators: Dr. John L. Sever
Mrs. R. Traub
Mrs. F. Moder
Mr. F. West

Cooperating Units: Dr. Margaret H. Jones
UCLA, Los Angeles, California

Man Years:

Total:	3
Professional:	1.5
Other:	1.5

Project Description:

Objectives: Use combined virus isolation and antibody determinations with serial specimens from pregnant women who register early in gestation as well as women with infertility and repeat abortions to determine the role of infections in these patients and their pregnancies.

Methods: An initial group of 5000 women were studied serologically for evidence of infection in relation to pregnancy outcome. This data is now being analyzed in detail.

At present serial sets of specimens for virus isolation and serology are collected for investigation of infertility and abortion. Cervical swabs, throat swabs, blood specimens and in some cases endometrial biopsies are obtained to study for infection.

Major Findings: The serological tests of the original 5000 patients have now been completed and are being analyzed. Several publications on toxoplasmosis and rubella have already appeared.

The studies of abortions and infertility have demonstrated infections with cytomegalovirus. Additional specimens are being collected and tested at present.

Significance of the Program to the Institute: The early registration of pregnant women in this population permits detailed studies of infections early in pregnancies and in well studied cases of infertility. The infections which occur early in pregnancy have not been investigated to any significant extent in the past because of the difficulty in obtaining these necessary specimens. These studies then provide an important opportunity to study infections at a critical period early in gestation.

Proposed Course of the Project: The analysis of the initial 5000 patient study will be completed in the next year. We will include IgM data during the year.

A special study of cytomegalovirus infections will be completed next year. This will provide longitudinal information on the occurrence of infections throughout pregnancy.

Specimens from women with infertility will be studied in detail. An additional 50 women will be included next year.

Honors and Awards: None

Publications: None

Contract # NIH-NINDS-69-4

Serial No. NDS (CF)-69 ID 1731
1. Infectious Diseases Branch
2. Section on Virology and Bacteriology
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Isolation of Infective Agents From Chronic Diseases

Previous Serial Number: Same

Principal Investigators: Dr. Luiz H. Barbosa, IDB, NINDS
Dr. David A. Fuccillo, IDB, NINDS
Dr. William T. London, IDB, NINDS
Dr. John L. Sever, IDB, NINDS
Dr. Monique Dubois, Postdoctoral Fellow, MS Society
Dr. Jerome Kurent, IDB, NINDS

Other Investigators: Mrs. Rebecca Hamilton, IDB, NINDS
Mr. Otto Gutenson, IDB, NINDS
Miss Helen Krebs, IDB, NINDS
Mrs. Anita Ley, IDB, NINDS
Mr. Leonard Moore, IDB, NINDS

Cooperating Units: University of Tennessee (Dr. J. T. Jabbour)
University of Vermont (Dr. George Schumacher)
Indiana University Medical Center (Dr. Wolfgang Zeman)
Wilmington General Hospital (Dr. George Boines)
Cornell University, N.Y. (Dr. Fred Plum)
Washington University, St. Louis (Dr. Mark May)

Man Years:

Total:	3.9
Professional:	1.4
Other:	2.5

Project Description:

Objectives: To establish whether persistent or tolerant viral infections are associated with chronic diseases such as Bell's palsy, amyotrophic lateral sclerosis, Creutzfeldt-Jakob disease, progressive multifocal leukoencephalopathy, and multiple sclerosis.

To purify and concentrate PML and SSPE viruses and examine these agents electron microscopically, biochemically, and antigenically in parallel with the conventional forms of these viruses.

To attempt the transmission of MS to SPE mice and chipanzees using intracerebral inoculation of 10% homogenate prepared with "plaque" areas from brain autopsy material.

To trace the possible anti-brain activity of MS immunoglobulin, using I^{131} labeled MS serum to inoculate Rhesus monkeys for brain autoradiography. To develop an animal model system to study the immunopathogenesis of SSPE as well as possible therapeutic approaches.

To conduct epidemiological survey concerning the occurrence of SSPE during the 1965-1975 decade.

To evaluate the role of cellular immunity, humoral immunity, and interferon in protecting experimental animals against neurotropic measles virus.

Methods Employed: Brain specimens from well-documented cases of MS, PML, ALS, SSPE, etc., were homogenized to a 10% suspension and inoculated intracranially in groups of 5 rhesus monkey fetuses during the first one-third of gestation when the animals are expected to be immunologically immature and hence more susceptible to infectious agents. These fetuses were carried to term and, immediately after birth, one animal was killed and examined histopathologically for CNS lesions. The remaining animals were carefully observed for clinical symptoms, abnormal behavior, and antibody pattern. A 2-year follow-up has been scheduled.

Brain tissue from patients with encephalopathies, muscle from patients with polymyositis, and lymph nodes from individuals with SSPE and MS were examined by electron microscopy, fluorescent microscopy and, whenever possible, cultured in vitro. These tissue cultures were submitted to virologic and serologic assays, inoculated into experimental animals, and cocultivated with human diploid and heteroploid cell lines in efforts to isolate and identify intracellular agents.

Brain autopsies from patients with PML and known to contain intranuclear virus were frozen and thawed three times, the nuclei extracted and disrupted by physical means. Nuclear extracts were isopycnically banded in CsCl and fractions examined under the electron microscope. Fractions containing the papovavirus were used to immunize guinea pigs in order to produce monospecific antisera intended to identify the virus. The peroxidase-coupled antibody technique was perfected for measles virus and it is expected to become a valuable tool when utilized with these guinea pig antisera and PML brain tissue. Similar approach, but using zonal centrifugation to purify the virus, was planned for SSPE and is awaiting approval of the necessary contract. The same technique is expected to be utilized to study particulate fractions of MS brain.

Comparative studies between SSPE measles and prototype measles virus were conducted in efforts to establish whether or not the SSPE isolates have markers to distinguish them from laboratory strains of measles.

A questionaire concerning the occurrence of SSPE since 1965 was sent to neurologists, pediatricians and pediatric neurologists at 104 medical centers throughout the USA. Data was analyzed in our laboratory and at the University of Tennessee.

In order to investigate a possible impairment of cellular immunity in SSPE patients, an in vitro lymphocyte assay was conceived and efforts initiated to obtain a measles chronically-infected cell line necessary to evaluate lymphocyte measles-specific delayed hypersensitivity.

Major Findings: Primary and secondary MS brain cultures were extensively tested for the possible presence of viruses. No evidence of an infectious agent was found in three MS brain cultures studied by immunofluorescence, electron microscopy, hemadsorption, cocultivation, and biochemical "shocking".

An immunoperoxidase electron microscopic test for measles virus was developed and used to study SSPE strains of the virus. This assay is expected to be useful on the search for antigens in MS, PML, and other neurological diseases. A papovavirus was purified from a PML brain autopsy and subsequently used to inoculate laboratory animals. The purification was achieved by banding the virus in CsCl (1.6 grs/ml). Spectrophotometry and electron microscopy monitoring resulted in a rather pure virus preparation of approximately 10^7 intact particles/5 grams of tissue.

Both PML tissue suspensions and the purified virus were utilized to inoculate a variety of tissue culture systems which, after months of careful follow-up, were considered free of PML virus as monitored by cellular changes and/or presence of intranuclear particles.

The search for biological markers to distinguish SSPE isolates from conventional measles virus proved unsuccessful. Marked differences between SSPE strains were noted by various parameters, indicating strain dissimilarities as great or even greater than those previously observed between laboratory strains of measles virus.

Epidemiological studies involving over 250 SSPE patients and dozens of medical institutions produced the following major findings: 1) the frequency of SSPE was approximately 1:1,000,000 during 1960-1970; 2) concentration of patients in the southeastern part of the USA; 3) the age of onset of the disease was 7.2 years; 4) a 3.3:1 male/female ration; and 5) early measles infection <3 years of age in the majority of patients.

Significance to the Program of the Institute: The development of mixed cell cultures to unmask latent infections provides an excellent methodology for the study of chronic diseases of possible viral etiologies.

The understanding of slow virus infections of the CNS will depend upon purification of the suppressed form of these agents followed by careful biochemical and biophysical analysis. To accomplish this goal, the use of zonal centrifugation for cellular fractionation should be emphasized.

Proposed Course of the Project: Special emphasis will be placed upon a brain culture research program. Investigation of the mechanism of pathogenesis and possible immune deficiencies in patients with neurological diseases will be conducted. Our selection of patients with multiple sclerosis, Parkinson's Disease, progressive multifocal leukoencephalopathy, and amyotrophic lateral sclerosis is supported by the existing data which suggest possible viral etiologies for each of these diseases. Tissue specimens and blood from patients will be provided through collaborative-contract arrangements with investigators throughout the country.

The possible viral etiology for MS will be exhaustively investigated. Since the pathology of Visna and MS are similar, and reverse transcriptase was found in tissues infected with Visna virus, we intend to determine whether this viral enzyme is present in spinal fluid and brain tissue of MS patients. In addition, molecular hybridization studies with MS tissue and tissue culture will be initiated to search for virus "fingerprints" such as messenger RNA, virus specific DNA or RNA. These homology assays would be conducted with MS purified DNA and RNA in presence of labelled measles RNA and possibly labelled varicella-zoster DNA. The latter viruses are at present the best candidates since serologic surveys indicated that MS patients have elevated measles and/or zoster antibody levels.

In a second approach, MS gamma globulins will be coupled to tracers (fluorescein and peroxidase) and used in a direct antigen-antibody test with MS brain tissue culture. Using the electron microscope and UV microscope, we can determine and identify antigenic sites in the MS brain tissue culture.

Third, in a more direct fashion, we will utilize experimental animals known to be surgically acceptable to slow neurotropic agents (scrapie, visna) to inoculate MS brain preparations. These animals will include SPF mice and primates.

Utilizing the mixed culture technique, we hope to determine whether it is possible to release suppressed virus from these chronic neurologic diseases and to gain a further understanding of the pathogenesis of latent infections of the CNS. Antibody levels and competence of lymphocytes from patients will be examined, using the standard techniques. The SSPE strain of measles virus will be studied in laboratory animals and efforts will be directed at the development of an animal model system for this disease.

Honors and Awards: None

Publications:

Price, R., Chernik, N. L., Horta-Barbosa, L., and Posner, J.: Herpes Simplex Encephalitis in an Anergic Patient. Am. J. Med. 54:222-228, February, 1973.

Serial No. NDS (CF)-69 ID 1732
1. Infectious Diseases Branch
2. Section on Virology and Bacteriology
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Investigation of the Role of Mycoplasma spp. in Perinatal and Neurological Diseases

Previous Serial Number: Same

Principal Investigators: Dr. David L. Madden
Dr. David A. Fuccillo
Dr. John L. Sever

Other Investigators: Mrs. Aurella Krezlewicz

Cooperating Units: D. C. General Hospital
National Naval Medical Hospital

Man Years:

Total:	1.00
Professional:	.25
Other:	.75

Project Description:

Objectives: To study the role of mycoplasma in diseases of man, particularly those associated with perinatal disease, infertility, or neurological disease. To develop animal models and to determine the pathogenesis of these diseases. To develop new tests for identification of mycoplasma.

Methods Employed: Vaginal swabs and amniotic fluid obtained prior to therapeutic abortions as well as small pieces of tissue from the resulting abortions were studied for presence of mycoplasmas. Positive cultures were identified by standard techniques. Immunoglobulin levels in the amniotic fluid were determined.

Urine samples and small pieces of vas deferens were collected from men undergoing vasectomy. These samples were cultured for presence of mycoplasma and positive cultures were identified by standard techniques.

Small pieces of biopsy material from a variety of neurological diseases and secondary tissue cultures are also being studied for presence of mycoplasma. These include such diseases as Subacute sclerosing panencephalitis, Alateral sclerosis, Multiple sclerosis, and Creutzfeldt-Jakob disease. These tissues are being cultured in standard mycoplasma media, and the positive isolated cultures are being identified by standard techniques.

Major Findings: Mycoplasma were not isolated from 75 amniotic fluids obtained during saline abortion procedures. Mycoplasmas were isolated from 12 of 18 abortions studied and from 22 of 30 vagina swabs obtained prior to abortion techniques. No detectable level of IgM was in any of the amniotic fluids. Detectable levels of IgG and IgA were found in the amniotic fluids. However, there was no correlation with the presence or absence of mycoplasmas.

Mycoplasma were not isolated from any of the vas deferens. Classical strains of Mycoplasma hominis were isolated from 7 of 98 urines and T-strain mycoplasma were isolated from 18 of the urines from men undergoing vasectomy. This study indicates that mycoplasma are not present in the vas deferens of normal males.

Significance to the Program of the Institute: A program devoted to studying the effects of mycoplasma in various diseases complements the virological studies currently being done. This study and its support given to other investigators may help to more accurately define the role of mycoplasma and other agents in disease.

Proposed Course of the Project: Further studies are being done on the association of mycoplasma in normal deliveries. Effects are underway to initiate a study to determine the role of mycoplasma in infertility and repeat abortion. Attempts to define the role of mycoplasma in neurological diseases will be continued. Continued efforts will be made to apply new techniques in the identification of mycoplasma and mycoplasmal diseases.

Awards and Honors: None

Publications:

Madden, D. L., Horton, R. E., McCullough, N. B.: Persistence of Mycoplasma Pneumoniae in Germfree Guinea Pigs. Proc. 23rd Annual Session, American Assoc. for Lab. Animal Science, 1972

Serial No. NDS (CF)-70 ID 1848
1. Infectious Diseases Branch
2. Section on Immunochemistry and Clinical Investigations
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Delayed Hypersensitivity in Chronic Viral Diseases

Previous Serial Number: Same

Principal Investigator: Dr. Earl B. Matthew, IDB, NINDS
Dr. John L. Sever, IDB, NINDS

Other Investigators: Dr. David A. Fuccillo, IDB, NINDS
Mrs. Mary Krasny, IDB, NINDS

Cooperating Units: None

Man Years:

Total:	.5
Professional:	.5
Other:	None

Project Description:

Objectives: To determine the role of delayed hypersensitivity (cellular immunity) in chronic viral infections.

Methods Employed: Macrophage migration tests (MI), lymphocyte transformation and lymphotoxin assay were used in this study.

Major Findings: Satisfactory tests for hypersensitivity for mumps and vaccinia viruses were developed. No satisfactory test could be developed for rubeola virus.

Significance of the Program to the Institute: Prior to the development of these tests, no satisfactory tests were available for measuring cellular immunity in humans to virus antigens. Unfortunately, some of the virus systems such as rubeola do not lend themselves to performance with these in vitro methods. Thus, for diseases such as subacute sclerosing panencephalitis and multiple sclerosis it is not possible to determine the cellular immune state of the patient for measles.

Proposed Course of the Project: This project has been terminated January 1, 1973. It has been changed to project NDS (CF)-72 ID 1982.

Honors and Awards: None

Publications: None

Serial No. NDS (CF)-70 ID 1849
1. Infectious Diseases Branch
2. Section on Immunochemistry and Clinical Investigations
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Chronic Infection with Cytomegaloviruses in Man and Animals

Previous Serial Number: Same

Principal Investigator: Dr. Donald Henson, IDB, NINDS

Other Investigators: Dr. John L. Sever, IDB, NINDS
Dr. David A. Fuccillo, IDB, NINDS

Cooperating Units: NIH, NCI
Dr. Edward Henderson
Dr. Ronald Yankee
Dr. Stuart Siegel
Dr. Arthur Levine
Armed Forces Institute of Pathology
Dr. A. J. Strano

Man Years:

Total:	2
Professional:	1
Other:	1

Project Description:

Objectives: This is a study of chronic cytomegalovirus infection antibody levels, virus excretions, lymphocyte response in diseases of man and mice. The investigation relates specific pathologic processes to antibody levels and virus excretion patterns.

Methods Employed: Chronic infection in the tissues of mice has been demonstrated. The response of the host depends on the anatomy of the organs infected.

Children with acute or chronic leukemia have been studied for approximately two years. Results indicate a correlation between clinical symptoms and rise in antibody titers to CMV.

Significance of the Program to the Institute: Cytomegalovirus causes congenital diseases and death as well as high rates of infection in normal pregnancies. It is also a complication of malignancies. Understanding of this chronic infection should be of great value in the prevention of treatment of the disease.

The Proposed Course of the Project: We have reported on the findings and this study was terminated January, 1973.

Honors and Awards: None

Publications:

Henson, D., Siegel, S. E., Fuccillo, D. A., Matthew, E., and Levine, A. S. Cytomegalovirus Infections During Acute Childhood Leukemia. J. Infec. Dis., vol. 126, no. 5, p. 469-481, 1972.

Serial No. NDS (CF)-71 ID 1903
1. Infectious Diseases Branch
2. Section on Virology and Bacteriology
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Investigation of the Etiology and Effect of Serum and Infectious Hepatitis in the Perinatal Period

Previous Serial Number: Same

Principal Investigators: Dr. David L. Madden, IDB, NINDS
Dr. John L. Sever, IDB, NINDS

Other Investigators: Dr. Benedict Nagler
Mrs. Betty Dunlop
Dr. Robert Purcell
Mrs. Mary Krasny

Cooperating Units: Lynchburg Training School and Hospital
Montgomery County Public Health Service
Abbott Laboratories
Electro-nucleonics

Man Years:

Total:	.75
Professional:	.25
Other:	0.5

Project Description:

Objectives: To determine the etiology of Australia antigen associated (serum) and infectious hepatitis. To determine thr relationship of hepatitis/congenital jaundice and postnatal jaundice. To develop animal models and new diagnostic tests for these diseases.

Methods Employed: A large epidemic of infectious hepatitis occurred in the Lynchburg Training School and Hospital during the summer of 1970. Serial samples of feces and serum were obtained from many patients prior to development of disease at the time of acute disease and post infection. Serum samples have been obtained from these patients at 6-, 12- and 18 months. Hepatitis B antigen and antibody determinations have been performed on all patients with Down's Syndrome on eleven wards and matched controls. In cooperation with Electro-nucleonics, Co., three serial samples of serum are being examined by electron microscopy and immuno electron microscopy

techniques for presence of Hepatitis A antigen.

Serum samples from young healthy teenagers who developed hepatitis following parenteral use of drugs and their immediate families have been collected and tested for hepatitis B antigen and antibody to determine whether hepatitis B antigen is spread by nonparenteral routes in normal family units. Selected samples from the perinatal collaborative project are being tested for presence of hepatitis B antigen and antibody.

Major Findings: Hepatitis B antigen was detected in 20% of the patients with Down's syndrome at Lynchburg Training School and Hospital and in 7% of the matched controls. Hepatitis B antibody was detected in 39% of the mongols and 57% of the non mongol matched controls. The total hepatitis B infection rate as measured by presence of either hepatitis B antigen or antibody was 56% in the mongols and 63% in the matched non-mongols. A direct correlation between exposure to antigen and occurrence of antibody was observed. A sex difference was also observed in that males had a higher frequency of chronic antigenemia carriers, lower antibody and in general more infection than did females.

In a study of six family units in Montgomery County in which a teenager developed hepatitis B, no nonparenteral spread of hepatitis B as measured by either presence of hepatitis B antigen or hepatitis B antibody seroconversion was detected in any of the exposed family members.

Significance to the Program of the Institute: Hepatitis in pregnancy has been associated with neonatal jaundice, Down's syndrome and other forms of mental retardation, among the institutionalized mentally retarded patients with trisomy 21 karyotypes have a high rate of chronic hepatitis B antigenemia. This study provides additional depth to determining the role of infectious agents in perinatal disease and mental retardation.

Proposed Course of the Project: Studies on the effect of hepatitis in a mentally retarded population are being completed. The determination of the various serotypes of hepatitis B antigen in a mentally retarded population is being pursued. The relationship between exposure to hepatitis B antigen and neonatal jaundice, hepatitis and mental retardation will be determined.

Awards and Honors: None

Publications: Dietzman, D.E., Madden, D.L., Sever, J.L., Landers, J.J. and Purcell, R.H.: Lack of Relationship Between Down's Syndrome in Newborn Infants and Maternal Exposure to Australia Antigen. Amer. J. Dis. Child. 124: 195-197, 1972.

Serial No. NDS (CF)-72 ID 1981
1. Infectious Diseases Branch
2. Section on Virology and Bacteriology
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Immunoglobulin M and Congenital Infections

Previous Serial Number: Same

Principal Investigators: Dr. Luiz H. Barbosa, IDB, NINDS
Dr. John L. Sever, IDB, NINDS

Other Investigations: Miss Helen Krebs
Mrs. Rebecca Hamilton
Mr. Otto Gutenson

Cooperating Units: University of Tennessee (Dr. Sheldon Korones)
Electro Nucleonics, Inc. (Dr. Edward Bond)

Man Years:

Total:	0.8
Professional:	0.2
Other:	0.6

Project Description:

Objectives: To study the antibody specificity of cord and newborn serum IgM as an investigational screening procedure to identify intrauterine or neonatal infections. High-risk pregnancies will be included in this study, using the elevated IgM level as index for possible perinatal infections.

Methods Employed: Part of this investigation is being studied in collaboration with Dr. Korones at the University of Tennessee. In these studies, serum specimens are obtained bi-weekly from newborn infants in the high risk nursery. Elevated IgM levels (>20 mg. %) will be used as indication of possible congenital or neonatal infection. Sera with high IgM content are then passed through prepared sepharose columns in order to separate the 7S from the 19S gamma globulins. Purified IgM samples are subsequently tested by the indirect immunofluorescent assay against CMV, HVH, Rubella, Toxoplasma, and Treponema Pallidum which are known to be the main causative agents of perinatal infections. Infected tissue cultures are used as the subs rate assay for the three viruses and toxoplasma and treponema smears are utilized for determining IgM antibody elicited by toxoplasmosis and syphilis, respectively. A fluorescein conjugated anti-IgM serum is employed to stain antigen-antibody complexes. A careful longitudinal follow-up of the children under study will be conducted by Dr. Korones to correlate

serological data with clinical observations and, when indicated, clinical specimens will be sent to the laboratories of the Infectious Diseases Branch.

A second part of this project involves the separation and antibody determination of the IgM in approximately 12,000 cord serum samples from babies with elevated IgM. These infants' samples are from the Collaborative Perinatal Study and are products of conception from mothers with serological findings indicating infections during pregnancy. Antibody specificity of the purified cord serum IgM samples are tested in the same manner described above.

Major Findings: The novel technique of antibody-coupled sepharose particles was perfected in collaboration with Dr. Bond (Electro-Nucleonics) and found to be useful to separate immunoglobulins from body fluids. This technique provides an unexpensive and rapid method to purify IgM in contrast with the elaborate, time-consuming preparative ultracentrifugation in sucrose gradients.

Measles specific IgM was found to be present in the serum of SSPE patients. This observation correlates with an early report by Connoly, et al., describing rubeola antibody in the IgM fraction of SSPE patient's spinal fluid.

Significance to the Program of the Institute: Several infectious agents are known to produce intrauterine infection and fetal damage. A great number of mentally retarded children are a consequence of congenital infection affecting the CNS. Cord and newborn serum IgM levels are usually elevated with congenital infections. Therefore, the quantitation and qualitation of cord and newborn serum IgM can be applied as an investigational screening procedure to identify high risk infants, or in studies of newborn in distress. More specific procedures can then be used to determine the cause for the elevation of this immunoglobulin.

Proposed Course of the Project: Comparison studies between specific immunofluorescence and latex agglutination techniques will be conducted to evaluate degree of sensitivity and specificity of cord IgM antibodies and candidate antigens (Toxoplasma, syphylis, CMV, rubella, and Herpes). Once the assay is perfected to a desirable reproducibility, approximately 1,800 high IgM and serum samples from the Collaborative Perinatal Research Study will be tested for specific IgM antibodies. The results will be correlated with the mother's serum antibody findings as well as mother's clinical history and the infant's findings.

Honors and Awards: None

Publications: None

Serial No. NDS (CF)-72 ID 1982
1. Infectious Diseases Branch
2. Section on Virology and Bacteriology
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Delayed Hypersensitivity in Chronic Viral Diseases

Previous Serial Number: Same

Principal Investigator: Dr. David A. Fuccillo, IDB, NINDS

Other Investigators: Mrs. Renee Traub, IDB, NINDS

Cooperating Units: Dr. Bellanti and Dr. Russell Steel
Georgetown University School of Medicine
Washington, D. C.
Mr. Monty Vincent
Microbiological Associates, Bethesda, Md.

Man Years:

Total:	1
Professional:	0.5
Other:	0.5

Project Description:

Objectives: To determine the role of delayed hypersensitivity (cellular immunity) in chronic viral infections.

Methods Employed: A microassay technique for cell mediated immunity was developed which will increase the number of specimens that can be done with the added advantage of requiring less blood.

A lymphotoxin assay to be used in this study for rubella has been developed. Other tests for CMV and Herpes are being worked on at the present time.

A rubella chronically infected cell line was found to be very useful for the lymphotoxin assay. This assay was developed and tested against immunized rubella children and normal results indicate a workable system is now available.

A rubeola chronically infected cell line was developed. This system is now being used in the study of SSPE patients to determine their cellular immune capabilities against this virus.

Significance to the Program of the Institute: Prior to the development of the above test, no reliable tests for measuring cellular immunity in humans to viral antigens existed. Antibody tests to viral antigens have been developed and used in this and other laboratories for many years. This represents a test for only part of the body's immune response. The development of lymphotoxin assay into a clinically useable test for virus infection now allows both forms of the body's protective immune mechanisms, humoral and cellular, to be tested simultaneously.

Proposed Course of the Project: To complete the above mentioned specific projects and to develop additional tests to elucidate how the cellular immune system is involved with other viral diseases, especially those that cause congenital infection. Congenitally infected infants excrete virus in the presence of high antibody titers, possibly indicating that their cellular immune system is impaired. The effect of complement as well as the function of cells other than lymphocytes can be studied with this system.

This test will also be used to determine the cellular immune capabilities of patients with MS and other neurological diseases.

Honors and Awards: None

Publications:

Steel, R. W., Hensen, S. A., Vincent, M. J., Fuccillo, D. A., and Bellanti, J. A.: A ^{51}Cr Microassay Technique for Cell-Mediated Immunity to Viruses. J. Immunology, 1973. (In press)

Serial No. NDS (CF)-72 ID 1983
1. Infectious Diseases Branch
2. Section on Virology and Bacteriology
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Chronic Viral Infections

Previous Serial Number: Same

Principal Investigator: Dr. David A. Fuccillo

Other Investigators: Dr. John L. Sever, IDB, NINDS
Dr. William T. London, IDB, NINDS
Dr. David L. Madden, IDB, NINDS
Dr. Luiz H. Barbosa, IDB, NINDS
Mrs. Flora Moder, IDB, NINDS
Mrs. Renee Traub, IDB, NINDS

Cooperating Units: Dr. L. Johnson
Harvard Medical School, Boston, Mass.
Dr. Mark May
McMillian Hospital, St. Louis, Mo.
Dr. Gary L. Gitnick
Dept. of Medicine, UCLA Center for Health Sciences,
Los Angeles, California
Dr. Donald Henson, NCI
Mr. Monty Vincent, Microbiological Association
Dr. Billingsley, Naval Medical Center, Bethesda, Md.

Man Years:

Total:	2.0
Professional:	1.0
Other:	1.0

Project Description:

Objectives: The main objective of this project is to establish the clinical and biological significance of the two different strains of herpes simplex virus (Type I, "oral" and Type II, "genital") cytomegalovirus, varicella, in the causation of human disease including carcinoma, ulcerative colitis, Bells' palsy, facial paralysis, herpes zoster and chronic neurological diseases.

Methods Employed: The principal methods employed are: (1) the indirect hemagglutination test, by which type specific herpesvirus antibody is identified; (2) mass serological surveys of materials from selected patients with special disease entities; (3) virus isolation, titration, and

characterization procedures; (4) monolayer cultivation of tissue with cocultivation; (5) fluorescent antibody studies of monolayer cultures for the presence of latent infection.

Major Findings: Patients with carcinoma in situ of the uterine cervix were demonstrated to have increased amounts of antibody to Type II herpesvirus. Antibody was also elevated in these patients two years before they developed cancer (collaboration with Dr. Johnson). CMV antibody in those same patients did not show a relationship to their disease.

In collaboration with Dr. Henson (NCI) cytomegalovirus (CMV) infections were studied longitudinally in 88 leukemic children; 24 patients had cultural evidence of CMV infection at some point. There appeared to be a good correlation between excretion of virus (in urine and/or throat) and presence of complement-fixing (CF) and indirect-hemagglutination (IHA) antibody; however, a few patients had detectable antibody without excretion of virus and vice versa. Compared with patients not excreting virus, patients shedding CMV had significantly more episodes of pneumonitis and fever with rash but did not have more episodes of hepatitis, fever of unknown origin, or upper respiratory-tract infections. Clinical syndromes attributed to CMV correlated with fourfold rises in CMV antibody titer only during hematologic remission. These data suggest that the urine and throat should be repeatedly cultured to detect infection with CMV. Moreover, to diagnose disseminated CMV infections in acute childhood leukemia, it may be necessary to repeatedly determine levels of antibody to CMV.

In collaboration with Dr. Gitnick, isolation of CMV was accomplished from biopsy specimens taken from ulcerative colitis patients. The etiology of ulcerative colitis is unknown, but this work may help determine the relationship of this virus to the pathogenesis of this disease.

Significance to Biomedical Research and the Program of the Institute: These investigations attempt to elucidate the pathogenesis of viral infections of the adult and fetus using immunological and virological techniques. Herpes simplex virus, CMV, and varicella have been agents which have received particular attention in these studies, since they have significant neurotropic capabilities in terms of newborn and adult encephalitides. There is considerable speculation that these viruses may have latent, "slow" virus potential in relationship to chronic diseases of humans, including carcinoma and central nervous system infection. Investigation of the clinical and biological properties of the two strains of herpes simplex virus have permitted more definitive establishment of such capabilities. Furthermore, the therapeutic potential of experimental drugs in animals with these infections may prove useful for future treatment of humans.

Proposed Course of Project: Expanded study of antibody and isolation of virus from women with carcinoma in situ and carcinoma of cervix in collaboration with Dr. Johnson. In collaboration with Dr. May we will serologically screen acute and convalescence serum from Bells' palsy patients for IgM antibodies to CMV, herpes zoster and herpes simplex. Attempts to culture agents from tissue and screen monolayer cultures by fluorescent antibody technique for latent virus.

In collaboration with Dr. Billingsley we will culture hysterectomy specimens for the presence of herpes, CMV and mycoplasma. This will help determine the focus of chronic infection and clinical importance in controlling infection to the fetus or newborn vasectomy specimens will also be cultured for the presence of virus inorder to determine if the male may be involved in the transmission of these viruses.

Honors and Awards: None

Publications:

Henson, D., Siegel, S. E., Fuccillo, D. A., Matthew, E., and Levine, A. S.: Cytomegalovirus Infections During Acute Childhood Leukemia. J. of Infec. Dis. Vol. 126, No. 5, Nov. 1972, pp. 469-481.

Farmer, G., Vincent, M. M., Fuccillo, D. A., Horta-Barbosa, L., Ritman, S., Sever, J. L., and Gitnick, G. L.: Viral Investigations in Ulcerative Colitis and Regional Enteritis. Am. J. Dig. Diseas. (In press).

Serial No. NDS (CF)-72 ID 1984
1. Infectious Diseases Branch
2. Section on Virology and Bacteriology
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Maternal Infection and Pregnancy Outcome

Previous Serial Number: Same

Principal Investigators: Dr. David A. Fuccillo, IDB, NINDS
Dr. John L. Sever, IDB, NINDS

Other Investigators: Dr. William T. London, IDB, NINDS
Mrs. Renee Traub, IDB, NINDS
Mrs. Mary Ruth Gilkeson, IDB, NINDS
Mrs. Flora Moder, IDB, NINDS

Cooperating Units: Pennsylvania Hospital (Dr. Corson and Dr. Bolognese)
Naval Medical Center, Bethesda, Md. (Dr. Billingsley)

Man Years:

Total:	4.5
Professional:	1.0
Other:	3.5

Project Description:

Objectives: Of the 3.5 million children born each year in the U. S. approximately 3% are mentally retarded. It appears that as many as 10% of these cases may be attributed to infectious disease. Our objective is to utilize various virological techniques in an intensive study of viruses to determine their role in the production of birth defects and related abnormalities. To develop virological techniques necessary for the investigation of the natural course of the disease as caused by the infectious agents.

Methods Employed: New virus isolation techniques and serum neutralization tests are used for large scale testing in the study of pregnant women and their children. The development of new techniques, such as the IHA test for Herpes hominis Types I and II and a new test for cytomegalovirus will now permit the determination of the frequency of virus experience among study populations along with the presence of antibody and change in antibody titer to the virus. These tests are being used to establish the reliability of other tests and to determine their sensitivity and specificity. A serologic study utilizing these tests on sera collected on the Collaborative Study population was conducted to determine the frequency of antibody changes

during pregnancy and the effect of these infections on the developing embryo. We have completed the testing of 2500 placentas for rubella virus for the purpose of identifying children with congenital rubella by the presence of rubella in the placental material. These reports have been prepared for publication.

The use of a fluorescent, specific anti-IgM test is proving to be a valuable method for identifying congenital infections. The use of affinity chromatography is also studied for its value in separation of IgM.

Rubella vaccine is being given to pregnant women negative for rubella (at the Pennsylvania Hospital) who are subsequently being aborted. This will help determine the effect of vaccine on the fetus.

Pregnant women will be studied for genital infection with herpesvirus and cytomegalovirus. The effects of virus on pregnancy during gestational period will be studied. Amniotic fluid will be taken during pregnancy to determine presence of virus and IgM determinations performed on the fluids to ascertain the presence of this globulin and its value in the early detection of in utero infection. Follow up on patients with herpes will be done for abnormal Pap smears and cancer of cervix.

Major Findings: Three new tests were developed for specific virus serology. These were hemagglutination tests for Herpes I, Herpes II, and Cytomegalovirus. The tests now provide highly specific, reliable, rapid, micromethods for virus serology with these agents. This is of particular value because previous methods were particularly laborious and expensive or not specific. Also, since these are hemagglutination tests, the anticomplementary effects found in many Perinatal Research Study sera can be avoided and the sera are now useable.

Virus isolation studies of women for herpesvirus show transient as well as persistent infections. Isolation rates have been rather low in the population studied so a new socio-economic group is now being studied along with a local population to eliminate possible transportation problems.

The fetal abortion studies, after rubella vaccinations, have been completed for about 75 cases and results have been reported.

The use of gamma globulin as prophylaxis against rubella-produced abnormalities was reported. A comprehensive review on viral teratology was prepared for publication.

Significance to the Program of the Institute: The results from these studies help determine what effect virus infection has on abnormal pregnancy outcomes and also provide valuable information on the epidemiological aspects of virus infections. The relationship of these in utero infections to various degrees of neurological dysfunction in later years of life is still unknown.

Proposed Course of the Project: Studies are now in progress on cytomegalovirus infections during pregnancy and in several population groups in Maryland. Additional studies are being conducted on Herpes Type I and II infections in women. Drug evaluation of Cytosine Arabinoside for CMV and 5-IDU for Herpes hominis is being conducted in monkeys. Experimental drugs are also being tested. Approximately 16,000 sera from abnormal pregnancy outcomes are being tested for Herpes I and II and CMV.

Honors and Awards: None

Publications:

Bolognese, R. J., Corson, S. L., Sever, J. L., Fuccillo, D. A., Lakoff, K. M., and Klein, J.: Rubella Vaccination During Pregnancy. Amer. J. Obstet. Gynec. 903-907, 1972.

McCallin, P. F., Fuccillo, D. A., Ley, A. C., Gilkeson, M. R., Traub, R. G., and Sever, J. L.: Gamma Globulin as Prophylaxis Against Rubella-Induced Congenital Anomalies. Amer. J. Obstet. Gynec. 185-189, Vol. 39, No. 2, Feb., 1972.

Newman, S. J., Fuccillo, D. A., Sever, J. L., London, W. T., and Mendez-Cashion, D.: A Serological and Epidemiological Study of Toxoplasmosis in Puerto Rican Children. Clinical Proceedings of Children's Hospital, 47-53, Vol. 28, No. 2, February, 1972.

Serial No. NDS (CF)-72 ID 1985
1. Infectious Diseases Branch
2. Section on Virology and Bacteriology
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Isolation and Identification of Viral Antigens and Antibody for Rapid Identification of Virus Strains and Diagnosis of Diseases

Previous Serial Number: Same

Principal Investigators: Dr. David L. Madden, IDB, NINDS

Other Investigators: Dr. David A. Fuccillo
Dr. Luiz H. Barbosa
Dr. Gabriel Castellano
Mrs. Mary Krasny
Mrs. Aurella Krezlewicz

Cooperating Units: Microbiological Associaties, Inc.

Man Years:

Total:	1.00
Professional:	.50
Other:	.50

Project Description:

Objectives: To isolate and identify several viral antigens from each of several viruses. To utilize these antigens for more specific, rapid, sensitive identification of antibody, and/or a more accurate identification of the viral agent.

Methods Employed: High-titered virus preparations have been prepared using tissue culture techniques. By use of ammonium sulphate precipitation, protamine sulphate, and streptomycin B precipitation, the viral proteins will be concentrated. The various classes of antigens will be separated and identified by use of gel filtration, polyacrylamide gel, electropheresis, and density gradient iso-electric focusing. These antigens will be inoculated into animals to produce specific antisera. Highly purified antigens and specific antibodies will be labelled with iodine 125 for radio-immunoprecipitation tests.

Major Findings: Application of precipitation, gel filtration and poly-acrylamide gel electrophoresis indicates that the HA activity of measles can be concentrated. However, because of the many cellular components of

approximately the same weight and size, it has been impossible to eliminate all of the non HA active measles proteins from the preparations. Labelling of the proteins present in the HA active fractions with Iodine I^{125} indicate that about 1/3 of the proteins are associated with HA activity. Applications of a sandwich type radio-immuno test for detection of measles antibody are in the rudimentary stages.

Efforts have been made to purify anti-herpes antibody from human antisera. The results are encouraging. However, high background and nonspecific absorption of labelled antibody must be eliminated before the sensitivity of the test is suitable for routine serological usage.

Significance of the Program to the Institute: The development of more specific antigens or antibodies which measure more accurately the immunological status of an individual is needed. (e.g. in predicting the outcome of pregnancy following exposure to known teratogenic agents.) Highly specific antigens or antibody may help identify the biological differences between nonpathogenic and pathogenic strains of organisms. In addition, they may help differentiate the biological differences between measles virus which usually occurs in classic Rubeola from what occurs in encephalitis and other neurological diseases such as SSPE.

Proposed Course of the Project: Further studies will be done to identify the antigens associated with the measles and rubella, Herpes I, Herpes II, and CMV viruses. Efforts will be made to produce highly specific antisera to each agent and to utilize the specific antigens and antibody for identification of viral agents.

Honors and Awards: None

Publications: None

Serial No. NDS (CF)-72 ID 1986
1. Infectious Diseases Branch
2. Section on Experimental Pathology
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Intrauterine Inoculation of Fetal Monkeys With Tissues From Patients With Chronic Diseases and Infections

Previous Serial Number: Same

Principal Investigators: Dr. William T. London, IDB, NINDS
Dr. Luiz H. Barbosa, IDB, NINDS

Other Investigators: Dr. David A. Fuccillo, IDB, NINDS
Dr. John L. Sever, IDB, NINDS
Mrs. Blanche Curfman, IDB, NINDS

Cooperating Units: None

Man Years:

Total:	1.70
Professional:	0.65
Other:	1.05

Project Description:

Objectives: The study of chronic diseases, such as Subacute Sclerosing Panencephalitis, Creutzfeldt-Jakob disease, Amyotrophic Lateral Sclerosis, Progressive Multifocal Leucoencephalitis, and Multiple Sclerosis following the inoculation intracerebrally of fetal rhesus monkeys.

Methods Employed: In many diseases, the fetus and young are more susceptible than the adult. The possibility of exploiting this greater susceptibility of the neonate was the basis of the design to inoculate the fetuses of pregnant monkeys at 100 days of gestation with CNS material taken from patients showing clincal signs of the above diseases. Animals are subsequently delivered by cesarean section and held in isolation chambers for 3 to 5 years. Animals are observed daily for abnormal signs. Sera and spinal fluid are collected for study every three months.

Major Findings: The inoculated animals are now 18-26 months of age, to date, no abnormal signs have been observed in any of the monkeys.

Surveys of sera and cerebrospinal fluid for elevated antibody titers to a battery of antigens revealed no differences between inoculated animals and controls.

Significance to the Program of the Institute: A short time ago, the hypothesis that chronic or subacute degenerative diseases of the nervous system might be transmissible would have been an unbelievable conception to most medically trained pathologists or neurologists. Todate, using animal models, at least two chronic degenerative conditions seen in humans have been transmitted. As these diseases become more fully understood, we may have new ideas for the study of other degenerative diseases of the brain and central nervous system.

Proposed Course of the Project: Will be continued along the perimeters outlined above.

Two animals from each disease group will be immunosuppressed with cyclophosphamide when they are 24-30 months of age in an attempt to activate any latent infection.

Honors and Awards: None

Publications: None

Serial No. NDS(CF)72ID1987
1. Infectious Diseases Branch
2. Section on Experimental Pathology
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Caloric and Protein Restriction in Pregnancy and Their Effect on Measurements and Biochemistry of Newborn Rhesus Monkeys.

Previous Serial Number: Same

Principal Investigators: Dr. William T. London, IDB, NINDS
Dr. Donald B. Cheek, Dept. of Pediatrics
Johns Hopkins University Medical School
Baltimore, Maryland

Other Investigators: Dr. John L. Sever, IDB, NINDS
Dr. Jonas Ellenberg, OB, NINDS
Dr. John G. Bieri, NIAMD
Mrs. Margaret Ashworth

Cooperating Units: Dr. William Nyhan
University of California - San Diego Branch
P.O. Box 109
LaJolla, California 92037

Dr. Ernest Bueding
School of Hygiene
Johns Hopkins Hospital
Baltimore, Maryland

Man Years:

Total:	0.95
Professional:	0.30
Others:	0.65

Project Description:

Objectives: Initiate a nutritional study of non-human primates using pregnant rhesus monkeys to test the hypothesis that there is no causal relation between maternal nutrition during pregnancy and certain sensory, pathological, immunological and biochemical characteristicis of the infant. It is considered that a monkey trial in which the concept of randomness is permitted is a necessary preliminary to a human trial.

Serial No. NDS(CF)72ID 1987
1. Infectious Diseases Branch
2. Section on Experimental Pathology
3. Bethesda, Maryland

Methods employed: In the nonhuman primate nutritional study, pregnant rhesus monkeys are maintained throughout pregnancy on one of four diets restricted in either calories or protein. One diet is deficient in both. The pregnant animals are delivered at 158 days of gestation by cesarean section and the infant's tissues are processed for biochemical analysis by the Hopkins Unit. The nutritionally deprived female monkeys are continued on their respective diets and studies for immunological responses to various antigens such as tuberculin, rubella virus and brucella vaccine (Strain 19).

Major Findings: Seventeen viable fetuses were delivered from monkeys maintained on one of 4 diets.

Group I - Control diet	(4 fetuses)
Group 2 - Protein deficient Calorie sufficient diet	(3 fetuses)
Group 3 - Calorie deficient Protein sufficient diet	(4 fetuses)
Group 4 - Calorie deficient Protein deficient diet	(6 fetuses)

All organs were weighed at sacrifice and anthropometric measurements taken. It was found that kidney development was retarded when protein deficiency was present in the maternal diet (groups 2 & 4). Tissues from fetuses in each group were assayed for various chemical determinants. No significant differences were found for any of the determinants between normal and deprived fetuses. This situation was influenced by 2 factors. (1) The numbers in each diet group were too small, (2) The level of protein deprivation was not low enough. The nutritionally deprived female monkeys did not exhibit any differences in their immunological responses to the various antigens.

Significance to the Program of the Institute: General obstetric practice in the United States is to restrict weight gain during pregnancy except where specifically contraindicated. Investigators analyzing the maternal weight gain/birthweight data from the Perinatal Research Program have concluded that this practice may be actively increasing the low birthweight rate, and possibly the perinatal mortality rate. Indeed, this practice may produce babies who are smaller than expected for gestation and who may be poor performers postnatally and of lower intelligence.

Proposed Course of the Project: Another group of pregnant monkeys (40) will be fed on diets which are more restricted in protein to extend stress on the CNS and hopefully magnify any CNS effects that may occur. If the preliminary examinations of CNS tissues of fetuses from these animals shows no significant differences between normal and deprived monkeys, this project will be terminated. However, if differences are found, our Ad Hoc review committee will be called upon to help with planning future studies.

Serial No. NDS (CF)-72 ID 1988
1. Infectious Diseases Branch
2. Section on Experimental Pathology
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Herpesvirus Induction of Cervical Cancer in Cebus Monkeys

Previous Serial Number: Same

Principal Investigators: Dr. William T. London, IDB, NINDS
Dr. Amos E. Palmer, IDB, NINDS
Dr. Andre J. Nahmias
Dept. of Pediatrics, Emory Univeristy
School of Medicine, Atlanta, Georgia
Dr. John L. Sever, IDB, NINDS

Other Investigators: Dr. David A. Fuccillo, IDB, NINDS
Mrs. Renee Traub, IDB, NINDS

Cooperating Units: Dr. John Verna
Meloy Laboratories
Springfield, Virginia

Man Years:

Total:	1.70
Professional:	0.70
Other:	1.00

Project Description:

Objectives: Several studies reported in literature have indicated that herpes simplex virus (HSV) Type II may be etiologically related to human cervical cancer. One of the important ways to demonstrate a causal relationship between congenital herpes and cervical cancer would be to determine if cervical cancer can be produced in monkeys inoculated genitally with the herpes simplex Type II Virus.

Methods Employed: We have shown earlier that, in various monkey species in which genital infection with HSV Type II was attempted, it was possible to infect only the Cebus monkey. One particularly interesting observation was that the Cebus monkey survived infection and that the infection mimicked closely with the clinical and laboratory findings which have been found with genital herpes infection in women. Since the first infection may not be sufficient to transform cells, and since reinfection can be established in the monkey, we are in the process of inoculating repeatedly 225 female Cebus

monkeys genitally with HSV Type II and 75 control female animals with non-infected tissue culture material.

Major Findings: A primary genital infection can be established in Cebus monkeys with the development of genital lesions in approximately half of the infected animals. The lesions occur on the vulva and on the cervix, with no neurological or visceral complications so that follow-up studies for the development of cancer are possible. As in humans, spontaneous genital recurrences can occur in infected monkeys. Reinfection was difficult to establish in recently infected animals. Venereal transmission from genitally infected female monkeys to males, and occasional viral transmission between female cage mates, possibly by way of a male has been observed.

The presence of herpes simplex virus (HSV) antibodies prior to HSV-2 genital inoculation does not appear to influence the rate of genital infection, as compared to animals with undetectable antibodies in their acute serum.

Significance to the Program of the Institute: The possible role of genital herpesvirus Type II infection in fetal and neonatal diseases, cervical cancer, and chronic neurological disease stimulated this research in nonhuman primates.

Proposed Course of the Project: Emphasis will be placed on establishing higher than 50% rate of infection in infected monkeys. The time of inoculation with relation to the menstrual cycle will be varied as well as modifications in the inoculation techniques to attain higher rates of infection. The natural infection of males will be followed clinically, virologically, and serologically. Several males will be infected by catheterization and the infection will be monitored in the same manner.

Honors and Awards: None

Publications: None

Serial No. NDS(CF)72ID 1989
1. Infectious Diseases Branch
2. Section on Experimental Pathology
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Transmission of Hepatitis B Virus to Sub-Human Primates

Previous Serial Number: New

Principal Investigators: Dr. William T. London, IDB, NINDS
Dr. Robert H. Purcell, IDB, NIAID

Other Investigators: Mrs. Blanche Curfman, IDB, NINDS

Cooperating Units: None

Man Years:

Total:	.65
Professional:	.25
Other:	.40

Project Description:

Objectives: The study and characterization of hepatitis B Virus in Rhesus Monkeys.

Methods Employed: A infectious pool of rhesus monkey-adapted, hepatitis B virus (HBV) will be inoculated in rhesus monkeys in order to do biological characterizations of the agent.

Major Findings: When the pool was administered by different routes, it was found that HBV was less infectious for the Rhesus monkey when administered orally than when inoculated parenterally. Rhesus monkey sera having HBAg was infectious for other rhesus monkeys. Whereas, sera containing HBAb was not infectious for monkeys. This demonstrates a close relationship between the detection of HBAg and the infectivity of the serum specimens tested. Early passages of HBV in rhesus monkeys resulted in a prominent antibody response with or without demonstrable HBAg. However, higher passages were characterized by transient development of HBAg but a diminished HBAb response. Serial liver biopsies obtained from rhesus monkeys prior to inoculation and throughout the incubation period, acute phase and convalescent period of infection with HBV were histologically normal. Furthermore, serum amino transferase levels did not become elevated during the HBV infection.

Significance to the Program of the Institute: This study developed as the result of serological surveys in the perinatal research program. With an animal model, the transmission and pathogenesis of the disease can more

easily be studied.

Proposed Course of the Project: The Rhesus monkey model will be used for further characterization of the agent of hepatitis B, such as: Effects of various disinfectant agents on HBV infectivity. What effects will banding in Cesium Chloride have on the virus? Will clinical signs of hepatitis be seen in monkeys immunosuppressed and then infected with HBV?

Honors and Awards: None

Publications: None

Serial No. NDS (CF) 72ID 1990
1. Infectious Diseases Branch
2. Section on Experimental Pathology
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Chronic Lead Poisoning in Rhesus Monkeys

Previous Serial Number: Same

Principal Investigators: Dr. William T. London, IDB, NINDS
Dr. Bernard C. Zook, D.V.M.
Assistant Prof. of Pathology
Department of Pathology
George Washington Univ. School of Medicine
Washington, D.C.

Other Investigators: Mrs. Margaret Ashworth

Cooperating Units: None

Man Years:

Total:	0.40
Professional:	0.10
Other:	0.30

Project Description:

Objectives: Establish the Rhesus monkey as an animal model for study of lead poisoning as it occurs in children.

Methods Employed: Adult and juvenile monkeys are given finely ground leaded paint orally via a stomach tube three times a week. Weekly doses range from 50 - 600 mg. lead/kg. of body weight. All animals have had base line hematology, urinalyses, urine delta-aminolevulinic acid, coproporphyrine III, serum vitamin B12 and blood lead tests. All tests are run at monthly intervals to monitor changes in these values. Radiographs of the right wrist have been taken each month of all immature monkeys.

Major Findings: Lead containing paint was administered orally to six adult, five juvenile and four infant rhesus monkeys for periods of 167-445 days. The dosages of lead required to produce toxic effects were considerably higher (200 to 600-fold) than has been estimated to be the toxic dose for children. Adult monkeys were more susceptible to the toxic affects of lead than juveniles, and juveniles appeared to be slightly more susceptible than infants. In no case were obvious neurologic signs observed in the

treated monkeys. We have found that the rhesus monkey is very resistant to lead poisoning, does not mimic the human condition and there fore would not be a ideal model.

Significance to the Program of the Institute: Lead intoxication continues to be a serious neurological hazard among children in the urban areas. An animal model is needed to study the dose-symptom relationships, since sequelae often develop even in the absence of overt symptoms. New therapeutic measures to remove excessive lead from the tissues from the onset of irreversible damage to the central nervous system can be more easily studied in the animal model than in children.

Proposed Course of the Project: This project will close as of June, 1973.

Honors and Awards: None

Publications: None

Serial No. NDS (CF)-72 ID 1991
1. Infectious Diseases Branch
2. Section on Virology and Bacteriology
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Immunologic Studies of Congenital Infections and Chronic Infections

Previous Serial Number: Same

Principal Investigators: Dr. Luiz H. Barbosa, IDB, NINDS
Dr. David A. Fuccillo, IDB, NINDS
Dr. William T. London, IDB, NINDS
Dr. John L. Sever, IDB, NINDS
Dr. Jerome Kurent, IDB, NINDS

Other Investigators: Mrs. Rebecca Hamilton, IDB, NINDS
Mr. Otto Gutenson, IDB, NINDS
Miss Helen Krebs, IDB, NINDS
Mr. Leonard Moore, IDB, NINDS
Mrs. Anita Ley, IDB, NINDS

Cooperating Units: Wilmington General Hospital (Dr. George Boines)
Georgetown University (Dr. Joe Bellanti)
University of Vermont (Dr. George Schumacher)

Man Years:

Total:	3.2
Professional:	0.9
Others:	2.3

Project Description:

Objectives: To conduct comparative study of the role of delayed hypersensitivity, humor immunity and interferon in neurotropic infections. To study the development of the fetal immune system. To develop immunologic methodology for evaluating intrauterine infections and CNS infections. To investigate the immunopathology of Multiple Sclerosis and other neurologic diseases. To conduct serologic survey with spinal fluid and sera from patients with chronic diseases of the CNS.

Methods Employed: Rhesus monkeys were inoculated in utero with various viruses at the first, second, and third periods of gestation. At appropriate intervals, the fetuses were removed and blood collected for immunological evaluation. Antibody levels were determined by CF tests; lymphocyte transformation was measured by uptake of tritiated thymidine in the presence of

virus antigens; interferon production was established by Sindbis HA inhibition assays. Concomitantly, the pathogenesis of mumps virus was studied in some fetuses to determine the fetal susceptibility at various stages of gestation and to correlate these observations with the immunological data.

Sera from patients with various chronic infections were selected for immunoglobulin analysis. The sera were precipitated by the ammonium sulfate method and concentrated 10 times in saline. Then preparations were ultracentrifuged in linear gradients of sucrose and 7S gamma globulins separated from 19S molecules. The purified immunoglobulin fractions were serologically evaluated against different antigens to determine antibody specificity of early and late immune response which might provide clues as to the nature of the causative agent. In addition, studies employing Sepharose columns were initiated to purify IgM from the sera prior to indirect immunofluorescent assays with specific anti-19S fluorescein-conjugated serum in tissue cultures infected with different agents. Since a constant residual amount of specific IgM seems to be associated with persistent viral infections and since the FA assay is the most sensitive serological procedure, it is conceivable that such approach could give us leads as to whether a given agent is associated with chronic infectious diseases.

Sera and spinal fluid from patients with MS were compared with specimens from patients with other neurological diseases using the immunodiffusion technique and neutralization tests. Highly concentrated measles virus antigens were utilized for the gel diffusion tests and the Edmonston strain of measles virus was employed for neutralization assays.

Lewis' rats were immunized with UV killed SSPE isolates and prototype measles virus combined with Freund's adjuvant and after antibodies developed and delayed hypersensitivity became manifest, these animals served as serum and lymphocyte donors to groups of histocompatible, non-immunized animals which were subsequently challenged with $10LD_{50}$ neurotropic measles virus.

Major Findings: Animal experiments indicated that great antigenic differences exist between the 5 SSPE isolates obtained in our laboratory. Some of the isolates were highly antigenic, eliciting extraordinary levels of HI and CF antibodies, whereas other strains proved to be poor antigens. Great variability was also found in their ability to induce delayed hypersensitivity.

Chronological evaluation of the interferon response of the fetal rhesus monkey following Chikungunya virus inoculation showed that in the first period of gestation the fetuses are uncapable of synthesizing interferon. In the second and third periods good levels of interferon were obtained.

Measles-specific IgM was detected in the sera of SSPE patients. The macroglobulin was separated in Sepharose column and its specificity determined by HI test. This findings suggested that the suppressed measles encountered in brains and lymph nodes of patients produces a continuous antigenic stimulus.

Neutralization test was found to be at least 16-fold more sensitive than both HI and CF in determining measles antibody values in CF and MS patients. This finding warrants the re-evaluation of rubeola titers in the CSF of MS patients, which, according to previous studies by HI and CF, have very low or trace values not encountered in the normal population.

Significance of the Program of the Institute: The successful development of a simple diagnostic test for SSPE now warrants further immunological studies with cerebrospinal fluid from patients with other neurological diseases in an effort to define antibody patterns of diagnostic value. Of utmost importance is the use of more sensitive (qualitative and quantitative) assays to study the increased gamma globulin levels encountered in the spinal fluid of MS patients. A careful serological investigation of the type and specificity of the immunoglobulins present in the brain of MS patients and appropriately matched controls could provide important clues to the possible infectious nature of the disease. On the other hand, an antibody mediated or autoimmune theory on the etiology of MS could also be investigated by virtue of purifying CSF gamma globulins followed by I^{131} labelling and inoculation into monkey brains. The animals would be killed at 8-hour intervals and autoradiography of the brain sections could conceivably show antibody-antigen reaction sites. Another approach would be a direct immunofluorescent assay using fluorescein-coupled CSF gamma globulin with MS brain tissue cultures.

Proposed Course of the Project: Special emphasis will be given to the study of the fetal immune system in an effort to understand the relationship between the maturation of the fetus immunological defenses and its resistance to transplacental infection. We will focus in the rhesus monkey model system as this non-human primate is known to be susceptible to intrauterine viral infections followed by severe malformations of the fetus.

Continued attempts to concentrate and identify specific gamma globulins from sera and spinal fluid of MS patients may prove of value in establishing whether or not an infectious agent is associated with MS.

Honors and Awards: None

Publications:

Barbosa, L. H., London,W. T., Hamilton, R., and Buckler, C.: Interferon Response of the Fetal Rhesus Monkey Following Viral Infection. Lancet (submitted).

Horta-Barbosa, L., Fuccillo, D. A., and Sever, J. L.: Viral and Protozoan Infections of the Newborn. In Resuscitation of the Newborn Infant, H. Abramson, Editor, C.V. Mosby Co. (in press).

Serial No. NDS (CF)-72 ID 1992
1. Infectious Diseases Branch
2. Section on Immunochemistry and Clinical Investigations
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: High Risk Pregnancies and Perinatal Infections

Previous Serial Number: Same

Principal Investigators: Dr. John L. Sever, IDB, NINDS
Dr. David A. Fuccillo, IDB, NINDS
Dr. Luiz H. Barbosa, IDB, NINDS
Dr. Sheldon Korones, University of Tennessee

Other Investigators: Dr. Gabriel Castellano, MBA

Cooperating Units: University of Tennessee

Man Years:

Total:	1
Professional:	.5
Other:	.5

Project Description:

Objectives: The purpose of this investigation is to study high risk pregnancies in relation to perinatal infections. To accomplish this, a special study was developed at the newborn nursery at the University of Tennessee. This is a high risk nursery where infants from the Memphis Metropolitan Area are brought for special newborn care and procedures. Serial blood specimens are obtained and tested for immunoglobulin levels as a means of identifying those children with possible intrauterine infection. In addition, other laboratory tests and specimens are obtained to further define the infections which may be present. This study focuses on a group of infants which is most likely to have difficulties in the newborn period and is most frequently involved with infections.

Methods Employed: Serial heel prick blood specimens are obtained during the first weeks of life. These specimens are tested for IgM levels using radial immunodiffusion techniques. Infants showing elevated IgM levels are then further tested using venous blood specimens, throat swabs, anal swabs and other tissue specimens. These latter specimens are forwarded to the laboratories of the Infectious Diseases Branch for study. Virus isolation procedures and specific antibody tests are used. The data is then analyzed in relation to the clinical findings in the child and treatment is instituted wherever appropriate.

Major Findings: Approximately 1000 children have been studied this year. We find that an unusually high rate of IgM elevation in this population. This is a new study and investigations are in progress to define the specific organisms which may be responsible for the congenital infections. One new organism recently identified in this population to be of significance is a trachoma like organism causing eye infection in the newborn period.

The Significance of the Program to the Institute: In our effort to identify the causes of neurological and other damage to the developing child; this population provides the unique opportunity of studying high risk pregnancies and thus developing a great deal of information with a minimal amount of study and testing. We are focusing in on a population which is at high risk for congenital infections.

Proposed Course of the Project: The study will continue for at least two additional years during which a total of 2500 patients will be studied. Combined serological and virus isolation techniques will be used.

Honors and Awards: None

Publications: None

Serial No. NDS (CF)-72 ID 1993
1. Perinatal Research Branch
2. Section on Immunochemistry and Clinical Investigations
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Amniotic Fluid, Antibody and Infection

Previous Serial Number: Same

Principal Investigators: Dr. John L. Sever, IDB, NINDS
Dr. Thomas Riechelfer, D. C. General Hospital

Other Investigators: Dr. David Madden, IDB, NINDS
Dr. David Fuccillo, IDB, NINDS

Cooperating Units: D. C. General Hospital, Washington, D. C.

Man Years:

Total:	1
Professional:	.5
Other:	.5

Project Description:

Objectives: This is an investigation of amniotic fluid obtained at therapeutic abortion or amniocentesis to determine immunoglobulin levels and to attempt to isolate infectious agents. To accomplish this, approximately 100 amniotic fluid specimens were obtained at the D. C. General Hospital. Early amniotic fluid specimens were obtained by suprapubic uterine taps in conjunction with therapeutic abortions. Later specimens were obtained usually in relation to determinations of intrauterine bilirubin. These specimens are then tested for IgG, IgA and IgM levels and are provided to our laboratories for virus isolation and mycoplasma studies.

Methods Employed: Amniotic fluid specimens were obtained at amniocentesis for therapeutic abortions or serial determinations of bilirubin levels. Specimens are immediately brought to the laboratory for IgM, IgG and IgA determinations using the radial diffusion method and counter current electrophoresis. Aliquots of the specimens were also given to our other laboratories for virus isolation and mycoplasma isolation. Serum specimens are available from the mothers for subsequent antibody determinations.

Major Findings: A total of 100 specimens have been studied. IgG has been found in almost all of these specimens. There have been no detectable

levels of IgA or IgM. There have been no virus isolations from any of the amniotic fluid specimens to date and mycoplasma studies are in progress. A number of fetal tissue specimens have shown mycoplasma.

Significance of the Program to the Institute: This is the first study of intrauterine immunoglobulin levels and the first attempt at isolation of infectious agents from the uterus. Previous studies have dealt with specimens obtained at term. This data is confusing both in terms of antibody levels and the presence of microorganisms since vaginal contamination can and does occur.

Proposed Course of the Project: We are in the process of completing this data for reporting during this next fiscal year.

Honors and Awards: None

Publications: None

Serial No. NDS (CF)-72 ID 1994
1. Infectious Diseases Branch
2. Section on Immunochemistry and Clinical Investigations
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Clinical Studies of Chronic Infections of the Central Nervous System

Previous Serial Number: Same

Principal Investigators: Dr. John L. Sever, IDB, NINDS
Dr. David A. Fuccillo, IDB, NINDS
Dr. Jonas Ellenberg, Office of Biometry, NINDS

Other Investigators: Dr. J. T. Jabbour, Memphis, Tennessee
Dr. George Schumacher, Vermont
Dr. Jacob Brody, EB, NINDS
Dr. Milton Alter, Minneapolis, Minn.
Dr. John Kurtzke, VA Hospital, Washington, D. C.
Dr. Luiz H. Barbosa, IDB, NINDS
Miss Helen Krebs, R. N., IDB, NINDS

Cooperating Units: University of Tennessee
Veterans Administration Hospital, Washington, D. C.
University of Vermont
University of Minnesota

Man Year:

Total:	2
Professional:	1
Other:	1

Project Description:

Objectives: Clinical studies are being conducted on chronic infections of the central nervous system. During this year, the investigations have primarily centered on subacute sclerosing panencephalitis and multiple sclerosis. These studies have epidemiological and therapeutic components. They involve collaboration of a number of groups through the United States.

Methods Employed: A registry for subacute sclerosing panencephalitis was initiated as a joint effort with Dr. J. T. Jabbour at the University of Tennessee. This registry now has data for more than 350 cases of SSPE. The reporting of patients is being continued to determine if there will be a change in the number of cases per year related to the widespread use of measles vaccine. This investigation has provided us with opportunities to study unusual patients

such as one individual with hypogammaglobulinemia, as well as SSPE in one of two identical twins. Additional studies have been conducted on the epidemiology of multiple sclerosis. Serum and spinal fluid specimens are being tested for antibody to a variety of viruses. Similar studies of Creutzfeldt-Jakob Disease and Progressive Multifocal Leukoencephalopathy are in progress.

Major Findings: SSPE has been found to be more prevalent in the southeastern part of the United States. In addition, more than half of the children have experienced measles in the first two years of life. The ratio of males to females is 3 to 1 with this disease. These observations suggest that early exposure to measles may be important in the pathogenesis of the majority of cases of SSPE. Other cases may be related to combined infections with other viruses along with measles. Studies of a child with X-linked hypogammaglobulinemia has shown that SSPE can develop without antibody to measles. Thus, the pathogenesis of this disease does not seem to be related to either antibody to measles nor antibody directed against the central nervous system. This has been important in our understanding of the mechanisms which produce this disease.

Serologic studies of patients with multiple sclerosis have consistently shown slightly, but consistently, elevated titers to measles among the patients when compared to matched controls. Antibody to other viruses have generally not been increased. The possible importance of measles in the pathogenesis of multiple sclerosis must be considered further.

Significance of the Program to the Institute: Clinical studies of SSPE, MS, and other chronic infections of the central nervous system permit direct investigation of the possible causes of these diseases, and provide us with an opportunity to study unique "experiments of nature" which often provide very valuable insight into the disease process. These studies are designed to take advantage of both the epidemiology as well as the direct laboratory approaches to the problems of chronic infections of the CNS.

Proposed Course of the Project: We plan to continue this SSPE registry to determine the effect of the widespread measles immunization program, which has been in effect in the United States for more than 10 years. If the vaccine influences the rate of SSPE, this change should occur in the next few years.

Our special studies of multiple sclerosis and other neurological diseases will utilize a combined laboratory approach with tissues as well as serum specimens.

Honors and Awards: None

Publications:

Brody, J. A., Detels, R., and Sever, J. L.: Measles-Antibody Titers Sibships of Patients with Subacute Sclerosing Panencephalitis and Controls. The Lancet, January 22, 1972, pp. 177-178.

Dietzman, D. E., Horta-Barbosa, L., Krebs, H. M., Madden, D. L., Fuccillo, D. A., and Sever, J. L.: Diagnosis of Subacute Sclerosing

Sever, J. L.: Subacute Sclerosing Panencephalitis Treatment. Science, Vol. 175, pp. 220-221. Jan. 14, 1972.

Whitaker, J. N., Sever, J. L., and Engel, W. K.: Subacute Sclerosing Panencephalitis in Only One of Identical Twins. New England Journal of Medicine, Vol. 287, 864-866. 1972.

Serial No. NDS (CF)-73 ID 2034
1. Infectious Diseases Branch
2. Section on Virology and Bacteriology
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Electron Microscope Immunoperoxidase Studies of Measles and SSPE Viruses

Previous Serial Number: None

Principal Investigators: Dr. M. Dubois-Dalcq, IDB, NINDS

Other Investigators: Dr. Luiz H. Barbosa, IDB, NINDS
Dr. David Fuccillo, IDB, NINDS
K. Worthington, IDB, NINDS
R. Hamilton, IDB, NINDS

Man Years:

Total:	1.3
Professional:	1.1
Other.	.2

Project Description:

Objectives: To develop a specific stain for viral antigen at the ultrastructural level.

To explore the differences between productive and non-productive measles infection by this technique.

Methods Employed: Different cell lines infected with SSPE strains are studied. Sera from SSPE patients with high measles antibody titer as well as normal human measles positive and negative sera are used. The antibody is coupled to the enzyme peroxidase in the direct method. In the indirect method, different sera are used and the label is obtained after treatment with an antihuman globulin coupled to peroxidase. The cells are incubated after fixation in glutaraldehyde or formaldehyde. The entire procedure including embedding is performed in situ in order to preserve the original cell to cell and virus to cell interaction.

Major Findings: In the productive measles infection, a strong specific stain was obtained on the surface of the infected cells and on the virus. At specific sites those sites were identified with greater precision than those reported by other investigators using the immunoferritin technique. Cellular antigen labeling was also obtained with the immunoperoxidase technique whereas this is

hardly possible with the immunoferritin technique. The reaction was intense on the "fuzzy" nucleocapsids whereas the "smooth" nucleocapsids were never labeled. This suggests a difference in their antigenic nature.

In the non-productive measles infection, a block in maturation of infectious virus at the cell membrane seemed to occur. Most of the nucleocapsids seen in the cells were of the smooth type and not labeled. In contrast, a specific stain could be detected on the infected cell membrane and the budding process was rare or absent. This suggests that the lack of fuzz around the nucleocapsids may be associated with failure of alignment under the modified membrane and the subsequent budding.

Significance of the Program to the Institute: The immunoperoxidase method allows good resolution of viral antigenic sites at high magnifications under EM and may be of a value in studies of the immunopathogenesis of SSPE and other chronic viral infections of the CNS, like Multiple Sclerosis. Indeed, intracellular viral components can be specifically labeled and detected even when conventional EM studies have not revealed the presence of virus. As the isolation of virus from chronic brain infections in humans encounters so many pitfalls, this technique might help to identify viral antigen in those tissues.

Proposed Course of the Project: The immunoperoxidase labeling technique will be further improved by using factors increasing the permeability of the cell membrane to the globulins.

The productive measles infection induced by SSPE measles virus will be compared to the one induced by the wild type of measles in tissue culture. The non-productive stage of measles infection will be further explored on two different cell lines. Brains of hamsters with SSPE and brain specimen from an SSPE case that died recently will be studied with the same technique.

Publications: None

Serial No. NDS (CF)-73 ID 2035
1. Infectious Diseases Branch
2. Section on Virology and Bacteriology
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Electron Microscopic Studies of Multiple Sclerosis

Previous Serial Number: None

Principal Investigators: Dr. M. Dubois-Dalcq, IDB, NINDS

Other Investigators: Dr. C. S. Raine, Albert Einstein College of Medicine, N. Y.
K. Worthington, IDB, NINDS

Man Years:

Total:	.5
Professional:	.4
Other:	.1

Project Description:

Objectives: To search for viral material under EM in an acute case of Multiple Sclerosis.

Methods Employed: Two hours after the patients death, slices of brain from plaque areas were fixed in glutaraldehyde and processed for EM.

Major Findings: This is the first EM study on an acute case of MS. Histological examinations revealed two types of lesions, one containing perivascular infiltrates of macrophages associated with ongoing demyelination and the other characterized by infiltrates of plasma cells, lymphocytes and macrophages frequently located in areas of complete demyelination.

Rare virus like material was found under EM in some parenchymal cells. This material resembles nucleocapsids of the myxovirus group but we do not yet have sufficient evidence to identify them with certainty.

Significance of the Program to the Institute: Multiple Sclerosis is one of the most widespread neurological diseases among young people in this country and its etiology is still unknown. Recent reports have suggested that MS may be associated with a viral infection. This may induce an autoimmune demyelination process.

Proposed Course of the Project: Further brain specimens will be studied if other patients die of the disease in Dr. Schumacher's Neurology Department. This material is rare and precious. Other EM studies are thus necessary to confirm or disprove the presence of viral material and to better understand the immunopathogenesis of the demyelinating areas (plaques). Also, the immunoperoxidase technique would help in the identification of the agent, by the use of different antisera against different viruses.

Publications:

None

Serial No. NDS (CF)-73 ID 2036
1. Infectious Diseases Branch
2. Section on Virology and Bacteriology
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Immunopathology of Chronic Neurologic Infections

Previous Serial Number: None

Principal Investigators: Dr. Luiz H. Barbosa
Dr. Jerome E. Kurent
Dr. Monique Dubois

Other Investigators: Mr. Otto Gutenson
Mr. Leonard Moore

Cooperating Units: None

Man Years:

Total:	0.9
Professional:	0.4
Other:	0.5

Project Description:

Objective: To study the immunopathology of SSPE-like illiness in hamsters. To establish the role of antibody and/or cellular immunity in the development of chronic neurotropic measles infection in the newborn hamsters. To evaluate the use of different chemotherapic agents on the treatment of SSPE-like syndrome of the hamster.

Methods Employed: Recent studies indicate that newborn hamsters with maternal antibody survive a lethal dose of HNT (Hamster Neurotropic measles virus) and, eventually a certain percentage of the animals develop a latent brain infection which resembles SSPE in man. The same phenomenon occurs with weaning hamsters, and it has been suggested that the presence of antibody associated with an immature cellular immunity is crucial for the production of the SSPE-like syndrome. This study proposes to manipulate these two components of the immune system in order to establish the roles of antibody and delayed hypersensitivity in the hamster model system.

Hyperimmunized animals served as donors of serum and splenocytes. Litters were divided into 4 groups of 5 litters each:

a) Virus alone
b) Virus and antibody

c) Virus and splenocytes
d) Virus and antibody and splenocytes

Conclusions will be drawn after a careful follow-up of all animals during which will be critical to distinguish between survivors with no evidence of virus and survivors undergoing suppressed measles brain infection (SSPE-like illness). This distinction must be done through immunofluorescence, H&E staining, EM and cocultivation.

The hamster model system is intended to be used to evaluate the effect of anti-viral drugs such as Ara-A, Ara-C, IUDR, and others.

Significance of the Program to the Institute: The understanding of the involvement of the immune system in the development of the newborn hamster SSPE-like illness is of critical importance. The application of this type of analysis will provide valuable information on the immunopathogenesis of chronic viral infections of the CNS and possible means of prevention and/or control of the disease in animals. These new insights into the causes of damage to the CNS will provide guidelines for new experimental therapeutic approaches for the human counterpart of the disease.

Proposed Course of Project: The combined immunologic and virologic study will continue for at least one more year during which we plan to evaluate our methodology and specific testing. Once they prove satisfactory a larger number of animals will be utilized in the experiments in order to give us solid statistical interpretation of results.

Honors and Awards: None

Publications: None

Serial No. NDS (CF) 73ID 2037
1. Infectious Diseases Branch
2. Section on Experimental Pathology
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Perinatal Carcinogenesis in Patas Monkeys

Previous Serial Number: None

Principal Investigators: Dr. William T. London, IDB, NINDS
Dr. Jerry Rice, EPB, NCI

Other Investigators: Mrs. Blanche Curfman, IDB, NINDS

Cooperating Units: Mr. William Rickman, Corbel Laboratories
Rockville, Maryland

Man Years:

Total:	.45
Professional:	.25
Other:	.20

Project Description:

Objectives: To establish a non-human primate model for chemical transplacental-perinatal Carcinogenesis. Viral agents will be used as possible co-Carcinogens.

Methods Employed: This project is an explanatory program to demonstrate transplacental chemical carcinogenesis in a non-human primate. The old world monkey, Erythrocebus patas is uniquely suited for this purpose because of its relative freedom from bacterial and latent viral infections. Particular emphasis will be placed on defining the periods during gestation of maximal sensitivity of different organ systems to the carcinogen 1-ethyl-1 nitrosourea (ENU). Parallels will be noted between both the morphological and clinical behavior of corresponding human tumors, and the results obtained in rodents with this carcinogen. Pharmacologic studies of the rates and extent of transplacental passage of this carcinogen in the fetal monkey will be made.

Major Findings: Since the project has been going for about six months, only preliminary studies of dosage have been made.

Significance to the Program of the Institute: Cancer is the third most frequent cause of death among infants and children in the United States and most industrialized countries. Many of the specific varieities of cancer encountered in children are found uniquely or predominately in this age group and not infrequently develop so early in life that a prenatal origin is either certain or highly probable. With the recognition of the association between exposure to diethylstilbestrol in utero and the development of vaginal adenocarcinoma during the second decade of life, the possible significance to human health of exposure to transplacental chemical carcinogens has become increasingly a matter of concern. Experimental studies on transplacental chemical carcinogenesis have been limited to rodent species, which differ significantly from man in many ways. Among these are the very much more rapid rates of fetal and neonatal development and maturation in rodents, a factor which may be responsible for the failure so far to observe any tumors strikingly different from those inducible in adults in rodents exposed in utero to potent chemical carcinogens. A non-human primate model would more closely mimic the human cases of transplacental Carcinogenesis.

Proposed Course of the Project: Will continue as outlined above. Time in gestation, when fetal tissues are most sensitive, will be correlated with maximal dosage. All newborn animals will be monitored for any indications of nesplasia.

Honors and Awards: None

Publications: None

Serial No. NDS (CF)-73 ID 2038
1. Infectious Diseases Branch
2. Section on Immunochemistry and Clinical Investigations
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Combined Clinical Viral and Immunological Investigations of Chronic and Subacute Disorders of the Central Nervous System

Previous Serial Number: None

Principal Investigators: Dr. Jerome Kurent
Dr. Luiz H. Barbosa
Dr. John L. Sever
Dr. David A. Fuccillo

Other Investigators: Mrs. Anita Ley
Mrs. Flora Moder

Cooperating Units: Medical Neurology Branch, NINDS, NIH
Dr. King Engel
University of Tennessee, Pediatric Neurology
Dr. J. T. Jabbour
University of Vermont, Dept. of Neurology
Dr. George Schumacher

Man Years:

Total:	1.0
Professional:	0.5
Other:	0.5

Project Description:

Objectives: To establish the possible viral etiology of multiple sclerosis (MS), amyotrophic lateral sclerosis (ALS), and Guillain-Barré syndrome and to determine the pathogenesis of chronic brain infection in the hamster as a model for subacute sclerosing panencephalitis (SSPE).

Methods Employed:

1) Seroepidemiology: Serum and CSF specimens from patients with MS, ALS, and Guillain-Barré syndrome and controls matched for age, sex, and race shall be studied. The presence of serum and CSF viral antibodies shall be determined by one of three methods depending upon the specific virus. These shall include indirect hemagglutination, complement fixation, and serum neutralization. Antigens shall include polio virus Types I, II, and III; varicella-zoster; herpes Type I and II; cytomegalovirus; mumps; rubeola; rubella; influenza A, coxsackie B3 and B4.

2) Immunofluorescence: The immunofluorescent technique shall be utilized in a search for viral antigen in cryostat-cut brain sections from MS and ALS. In addition, ALS serum shall be studied for the presence of activity against viral antigen in CNS tissue sections from mice with motor neuron disease associated with C-type virus particles. Similarly, mouse serum activity against ALS tissue shall be studied with the immunofluorescent technique in an attempt to demonstrate immunological cross reaction between these two types of motor neuron disorders.

3) Tissue Culture: Muscle biopsy specimens from five ALS patients shall be grown as explant cultures and observed carefully for possible cytopathic effects. Immunofluorescent assays shall be conducted with patient's serum and if indicated with pooled ALS sera and normal serum. Other tests to detect the presence of an unconventional virus will include interferon assays with supernatant fluids of the cultures, hemadsorption tests with human Type O, monkey, guinea pig, and sheep erythrocytes, and a test for the presence of intrinsic interference by superinfection of cultures with Coxsackie A9. Attempts to rescue defective virus will be carried out according to three basic techniques; 1) cocultivation of the muscle cells with WI38, HeLa, and human spongioblast; 2) "shocking" of the muscle cultures with 5-iododeoxyuridine; and 3) forced fusion of monolayers with lysolecithin.

4) Experimental SSPE in Hamsters: Evaluation of the importance of a deficient or immature immune system in the pathogenesis of chronic viral encephalitis shall be undertaken utilizing a hamster model for SSPE. Suckling hamsters will be treated with either immune serum, immune splenocytes, or both, while receiving intracerebral measles virus. It is expected that this treatment shall render some degree of protection from acute fatal encephalitis, and that a proportion of protected animals shall develop chronic encephalitis. Depending upon which group(s) survives, conclusions will be drawn regarding relative importance of the immune system in the production of chronic viral encephalitis.

Major Findings: These studies are currently in progress.

Significance of the Program to the Institute: The search for viral etiologies of chronic and subacute human CNS disease by means of immunologic and virologic techniques is in keeping with the major objectives of the Infectious Diseases Branch. Implication of a virus in the etiology of these disorders would help provide the rationale for their prevention and treatment, as well as to set the stage for an understanding of the pathogenesis of chronic neurological disease.

Proposed Course of Project: If serological investigation of MS, ALS and Guillain-Barré serum and CSF specimens reveals as association between a particular virus and either of these disorders, more sophisticated efforts could be made to establish an etiological relationship. Tissue culture approaches utilizing conventional techniques as well as explant cultures of diseased neural tissues would be employed. Special procedures to allow defective viral agents to express their infectivity would also be utilized as described with ALS muscle biopsies.

If results of chronic CNS infection by measles virus in the hamster suggest a specific role for a compromised immunologic response in the pathogenesis of chronic viral encephalitis, attempts to adapt this model to a primate system would be made. Antiviral chemotherapy could be instituted in chronically infected animals, and favorable results possibly applied to the treatment of SSPE in man.

Honors and Awards: None

Publications: None

Serial No. NDS (CF)-73 ID 2039
1. Infectious Diseases Branch
2. Section on Immunochemistry and Clinical Investigations
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Blighted Potatoes and Birth Defects

Previous Serial Number: None

Principal Investigators: Dr. Jerome Kurent, IDB, NINDS
Dr. John L. Sever, IDB, NINDS
Dr. William T. London, IDB, NINDS

Other Investigators: Dr. Ken Deahl

Cooperating Units: Dr. Steve Sinden
Dr. Ramon Webb
Plant Genetics and Germ Plasm Institute
Vegetable Laboratory
Range 1 Agricultural Research Center West
Beltsville, Md. 20705

Man Years:

Total:	0.4
Professional:	0.2
Other:	0.2

Project Description:

Objectives: To determine if a relationship exists between maternal contact with blighted potatoes and the production of two severe birth defects of the central nervous system, anencephaly and spina bifida.

Methods Employed: The approach to this problem is two-fold:
(1) Animal feeding studies utilizing blighted potatoes (potatoes infected with the fungus of late blight, Phytophthora infestans) and swine, and
(2) Analysis of prospectively collected case reports of anencephaly and spina bifida from the C&FR data.

Major Findings: This investigation is now in its preliminary stages.

Significance to the Program of the Institute: Results of this study shall hopefully support the hypothesized etiologic relationship between maternal contact with blighted potatoes and the production of severe birth defects. This would help to establish guidelines for potato avoidance in order to reduce the incidence of these defects, and would also establish the teratogenic potential of blighted potatoes for the central nervous system.

Proposed Course of the Project: If birth defects are induced in swine following maternal feeding of blighted potatoes, studies utilizing primates would be undertaken, and efforts made to isolate and characterize the teratogenic compound (s).

Honors and Awards: None

Publications: None

ANNUAL REPORT
For Period July 1, 1972 through June 30, 1973
Office of Biometry

Collaborative and Field Research Program, National Institute of Neurological Diseases and Stroke

The Office of Biometry is a Branch within the Collaborative and Field Research Program, NINDS, and operates under the direction of the Associate Director, C&FR. The Office of Biometry is a central resource to NINDS for service in statistics, mathematics, computer technology, systems analysis and data processing. Its services range from brief consultations to participation as co-investigators on major projects.

The Office of Biometry carries out research in mathematical statistics for current and anticipated use in NINDS programs. It also provides consultation and service to other organizations undertaking studies in the area of NINDS interest.

The Office of Biometry consists of the Office of the Chief, the Section on Mathematical Statistics, the Section on Experimental Statistics, the Section on Applied Statistics, and the Section on Systems Design and Data Processing.

The report on the projects and services performed by the Office of Biometry is listed by major Program area, and followed by brief descriptions of Research, Miscellaneous Activities, Publications, and Future Plans.

OFFICE OF THE DIRECTOR, NINDS

1. Automatic Data Processing (ADP) Evaluation

A staff member of the Office of Biometry continues to serve as Chairman of the ADP Evaluation Task Force of the Institute, which includes other members of the Office. This is the coordinating and advisory group to the Branches and Laboratories which use ADP, for updating their five-year estimate of financial needs, for current inventory of equipment, and for other program information concerning data processing, for the HEW annual ADP report.

COLLABORATIVE AND FIELD RESEARCH PROGRAM

Office of the Associate Director, C&FR

1. NINDS Reorganization Plan

The Office of Biometry participated in the development of a comprehensive reorganization plan of the NINDS.

The plan was developed after detailed reviews of the organizations and reorganizations of the sister Institutes.

The objectives of the reorganization plan were to develop an organization which would emphasize planning, focus on priorities to achieve long-range goals, permit flexibility as priorities shifted, and provide a balanced program of directed and basic research.

Specific assignments of the Office of Biometry were to delineate the NINDS research activity process, as well as to design the major elements of an Office of Program Planning and Evaluation.

2. Presentation of the C&FR Program

The Office of Biometry participated in the development of a two-hour program presentation which described the C&FR Program objectives.

The C&FR Program was presented to the Acting Director, NINDS, and, after further revision, to the NANDS Council.

3. Progress report on the Collaborative Perinatal Program (CPP) to serve as background for publicity

At the request of the Associate Director, C&FR, the Office of Biometry prepared a report which described the highlights of past research of the CPP, and of the major studies that will be accomplished as part of the CPP comprehensive plan for data analysis.

The report provided part of the basis for an article on the CPP published in the Washington Post.

4. Cerebral Death Study

Principal Investigator: Dr. G. Molinari

At the request of the Associate Director, C&FR, the Office of Biometry reviewed a series of progress reports on the Cerebral Death Study, and observed meetings of the Principal Investigators and their Advisory Committee concerning the review of the pilot study, and plans for the major phase.

The Office of Biometry gave a presentation to the Associate Director, C&FR, and selected members of his staff, which summarized the pilot phase data, provided probability statements on the risks of patients surviving the various criteria of death, and estimated the numbers of patients required for a main phase.

Perinatal Research Branch

The Office of Biometry provides extensive support to the Collaborative Perinatal Project (CPP).

It participates in the Coordinating Committee for Data Analysis, an advisory group with expertise in the various disciplines associated with the CPP; it has participated in the development of a major comprehensive plan for data analysis, and routinely reviews proposals for studies of the CPP data.

The Office of Biometry also participates in the Epidemiologic and Statistical Advisory Committee. This group provides review and advice on statistical and epidemiologic problems.

The Office of Biometry participates in the special study groups charged with the development, analysis, and reporting of major studies of the CPP.

The assignment of personnel is as follows:

Statistician	Study Groups
Dr. Ellenberg	Cerebral Palsy, Congenital Malformations
Dr. Shaughnessy	Mental Retardation, Learning Disorders
Mr. Rubinstein	Communicative Disorders
Dr. McDonagh	Visual Abnormality, Convulsive Disorders
Dr. Chen	Minimal Brain Dysfunction, Pathology
Miss Jackson	Prematurity

1. Minimal Brain Dysfunction (MBD) Study

Principal Investigator: Dr. B. Fox

Initially, the participating statistician spent a considerable amount of time in reviewing existing literature of MBD and in familiarizing himself with the Collaborative Perinatal Program (CPP) data system.

A study plan has been developed by the principal investigator with his consultation. In Phase I, the study effort concentrates on defining various types of minimal brain dysfunction based on all related psychological and neurological data available from the CPP file. Toward this end, an extended list of relevant variables is being developed. Preliminary data processing requests have been made to derive normal limits for certain variables such as the discrepancy between verbal and performance IQ scores.

These variables will be checked further by psychological and neurological consultants for their symptomatological meaning relevant to MBD. The variables found relevant will then be subject to extensive statistical investigation; they will be examined with respect to their distributions, correlation clusters, and other individual and joint statistical properties. From these analyses, certain interpretative clusters of the variables under study may be found to describe various types of MBD.

Phase II of the study will relate prenatal, birth and delivery, neonatal or some postnatal variables, etc., to the MBD variables.

2. Communicative Disorders Study

Principal Investigator: Dr. P. La Benz

The Office of Biometry prepared the structure and detailed layout of extensive tabulations of the 8-Year Speech, Language, and Hearing (SLH) data. It also participated in the preliminary formulation of indices relevant to the SLH area with particular emphasis on neurological aspects. Currently, the work centers on the detailed implementation of the initial plans. Included are the search for suitable correlation coefficients and for appropriate clustering techniques.

3. Study of Mental Retardation at Age Seven

Principal Investigator: Dr. S. Broman

Detailed study plans have been developed from the general objectives listed in the comprehensive plan for data analysis. Outlines of statistical analyses necessary to accomplish the objectives have been prepared. Requests for frequency distributions necessary to the early stages of planning are being processed. Developmental work is nearly complete on a data processing request for creation of a subsample of cases. The subsample will allow the investigators to examine most of the variables for their relevance to mental retardation and to develop and refine statistical methodology prior to the completion of the data collection in early 1974.

4. Study of Learning Disorders at Age Seven

Principal Investigator: Dr. S. Broman

Progress in this area parallels that made in the above-mentioned area of mental retardation. Since many of the same variables must be analyzed for both studies, the same subsample has been designed to incorporate all variables of known or anticipated importance in the areas of mental retardation and learning disorders.

5. Cerebral Palsy Study

Principal Investigator: Dr. K. Nelson

A five-member study group has begun the task of analysis of the CPP data. The problem of defining Cerebral Palsy has been given the highest priority. Then, studies to identify antecedent etiologic factors of CP outcomes will follow.

The Office of Biometry has contributed to the design of the proposed analyses. Several possible methological approaches are being investigated, such as 1:1 matched pairing of CP cases with non-CP cases; comparison of CP cases with the non-afflicted portion of the population, with a stratification procedure removing confounding or nuisance factors.

6. Convulsive Disorders Study

Principal Investigator: Dr. K. Nelson

Initial planning involves classification of convulsive disorder cases. The first step is to establish seizure classes from the CPP data and to then investigate the spectrum of variables of each class. Examination of the pertinent data is underway, as well as the selection of appropriate statistical methods to achieve the above objectives.

7. Birthweight-Gestation Study

Principal Investigator: Dr. J. Hardy

The Office of Biometry is represented on this study group which has developed a proposal for a study of prematurity based on the CPP data.

An orderly progression of analyses has been developed to provide insight into the characteristics, causes, and consequences of prematurity.

These analyses concentrate on birthweight and gestational age, and their combined effects. Other measures such as placental weight, will be investigated to determine whether their addition to the combination will more precisely define prematurity.

New indices of risk, based on combinations of these 'prematurity' variables, will be created and tested to determine their utility in assessing the etiology of prematurity.

Cross-tabulations and sophisticated methods of statistical multivariate analysis will be employed.

8. Maternal Infection Study

Principal Investigator: Dr. J. Sever

The sera of mothers of children with specified abnormalities are being compared with the sera of matched mothers in search of relationships between the incidence of abnormalities and serologically inferred infections during pregnancy. The Office of Biometry is continuing its support of this study. Matching procedures have been implemented; a set of simple statistical tests has been devised; these have been prepared for detailed computer specification writing which will include an effective format for data display. Some early and therefore limited serology data has been analyzed.

9. Convulsions During Childhood Study

Principal Investigator: Dr. K. Nelson

Dr. Nelson selected a cohort of children who experienced a seizure during the first year of life. These children were to be studied with regard to their subsequent seizures and their mental development. The Office of Biometry participated in the analysis of the study. Various informative tabulations were prepared, and t tests, chi-square tests, and sign tests were made. Among other results, afebrile seizures and seizures with simultaneous dehydration were found to be the strongest prognosticators of later deficits. These results were reported at a meeting of the Child Neurology Society.

10. Preparation of a book on the CPP, "The First Year of Life"

"The First Year of Life", the second volume of a continuing series of books on the basic data of the CPP, is in preparation.

A member of the Office of Biometry is coordinating this major study, which includes the preparation of additional requests for data, their analyses, the procurement of computerized graphics and the preparation and review of the text of the book.

11. Growth and Development Study

Principal Investigator: Dr. G. Bartlett

A member of the Office of Biometry reviewed the manuscript of the monograph "Physical Growth from Birth to Seven Years" by Dr. Bartlett. Significant suggestions were made for improvement. An analysis of covariance for estimating secular trends and the effect of the socio-economic index on growth variables was recommended and specifications for its use were provided.

12. Computer Package for Multivariate Analyses

A member of the Office of Biometry collaborated with the Section for Production of Data Analyses, PRB, on the development of a series of programs for prospective use in the CPP. Programs for efficient computations of correlation coefficients had first priority. Three problems were studied; missing observations, large data base, utilization of computations relating to small groups in computations relating to more inclusive groups. The end product of these programs will be an analysis of covariance which will incorporate conventional regression analysis.

13. Interactive Statistical Program System to Facilitate Multivariate Analyses

In order to facilitate the use of large-scale statistical analysis techniques for the CPP, the Office of Biometry is in the process of developing a multivariate program system. The basic programs allow the user to compute correlation matrices for input into various multivariate procedures. The entire program system is designed to be interactive, that is, the user can communicate with the computer during execution via the program. The major cost reducing component of this system would be the elimination of numerous recalculations of essentially the same correlation matrix for different kinds of multivariate analyses. The major time-saving component is, of course, the very rapid turn-around associated with time-shared computer processing.

14. Georgetown University Retrospective Mental Retardation Study

Principal Investigator: Dr. Z. Shakhashiri

The Office of Biometry provided statistical consultation and service in the analysis of the Georgetown University Retrospective Mental Retardation Study. Mentally retarded children were compared with control siblings if available, and separately with non-sibling controls. The comparisons were made by chi-square and/or t tests.

15. Joint Frequency Analysis of Dichotomized Data

Certain data of the CPP consist of binomial scores, or multinomial scores, which can be reduced to a number of binomial scores. The quantity of data is large, involving about 28,000 or more cases and often 30 or more variables are used. An efficient method was developed to compute frequencies of various outcomes of interest. The computer approach involves sorting and counting of binary bits.

Infectious Diseases Branch

1. Analysis of Chord IgM Levels from Births in the CPP Study

Principal Investigator: Dr. J. Sever

The goals of this study are the examination of the association of chord IgM levels with specific mother and/or child abnormalities, and etiologic characteristics of the abnormal group. The Office of Biometry has collaborated on the design and analysis of the study. A normal IgM distribution, stratified for key relevant variables such as sex, race and maternal age was developed. The next step will be a comparison of the chord IgM distributions for abnormal groups with that of the normal IgM distribution, accounting for as many related factors as sample size will allow in a multidimensional fashion. The statistical analyses of these data will be presented at the 4th International Conference on Birth Defects and published subsequently in a monograph co-authored by a member of the Office of Biometry.

2. Maternal Clinical Infections and Pregnancy Outcomes Study

Principal Investigator: Dr. J. Sever

Clinical infections during pregnancy were reported in approximately 5000 cases of the CPP. The children have been followed for 5 to 8 years. Data for all the blood samples of the infected mothers will be analyzed in relation to pregnancy outcome. The Office of Biometry has collaborated on the design and analysis of this study. Basically the infected mothers will be matched with non-infected controls and comparisons made of rates of specified outcomes. The effects of variables which have not been accounted for in the matching procedure, will be evaluated. A preliminary report will be given at the 4th International Conference on Birth Defects and published subsequently in a monograph with a member of the Office of Biometry as a co-author.

3. Primate Information Retrieval System

Principal Investigator: Dr. W. London

The Office of Biometry has completed a major study which is changing and modifying the data storage and record keeping procedures for primates used by the Infectious Diseases Branch. In addition,a "Primate Information Retrieval System" has been developed as a computer application which now permits additional information to be retrieved. The systems design, documentation and specifications for this system have been completed, and

programming of the more than 40 retrieval programs and the several file structure programs is underway. The projected completion date is June, 1973.

4. Primate Maternal Flu Infection and Hydrocephaly Study in Neonates

Principal Investigator: Dr. W. London

Office of Biometry staff is currently involved in the planning of a series of animal experiments to examine the possible associative relationship of maternal flu infection with hydrocephaly in the newborn. The studies are designed to demonstrate any real association, and to generate information dealing with the rate and site of infection as determinants of the degree of hydrocephaly. Our primary responsibility to date has been the statistical design of the experiment and the determination of appropriate sample sizes.

5. Cebus Monkey Normal Blood Level Study

Principal Investigator: Dr. W. London

This study is designed to establish normal hematologic values for the Cebus monkey. A member of the Office of Biometry staff participated in the planning and design of the study. Collection of blood specimens is continuing on a large pool of Cebus monkeys. Office of Biometry personnel will be responsible for analysis of data, which will include graphical presentation of the hematologic data and establishment of statistical limits or bounds on what will be used as a baseline for future studies involving the Cebus.

6. Primate Maternal Nutrition Study

Principal Investigator: Dr. W. London

This project involves the study of caloric and/or protein restriction in pregnancy and its effect on biochemistry, and neurological deficits of newborn Rhesus monkeys. The pilot study has been completed, with 22 out of 24 monkeys successfully impregnated and maintained on their specified diets. Approximately 600 measurements relating to the newborn and the mother were analyzed for presentation to an ad hoc review committee, convened to consider continuance of the study. Two-way analyses of variance were used for all variables, with a full least squares approach taking account of the unequal number of animals in the cells.

The Office of Biometry provided systems operation, data processing support, and computer graphics output.

A manuscript is in preparation for the Journal of Nutrition, which will deal primarily with the many previously uninvestigated areas of research in the methodology of primate nutrition. A member of the Office of Biometry is a co-author of this future publication.

7. Herpesvirus Induction of Cervical Cancer in Cebus Monkeys

Principal Investigator: Dr. W. London

Several studies have indicated that herpesvirus (HSV) type 2 may be etiologically related to human cervical cancer. This study is investigating the possible causal relationship between genital herpes and cervical cancer by attempting to induce cervical cancer in monkeys inoculated genitally with the virus. The statistical design of the study was produced by the Office of Biometry in FY 72. Continual monitoring of the statistical aspects of the study design have been necessary due to the development of heretofore unknown conditions relating to the interaction of the HSV-2 virus in the Cebus model.

A manuscript has been submitted for publication to the Journal of the National Cancer Institute, relating the methodological approach to the problem. A member of the Office of Biometry Staff is a co-author.

Epidemiology Branch

1. Phase I Analysis of Geographic Distribution of CVD and MS Death Rates

Principal Investigator: Dr. D. Reed

Office of Biometry participation in this project for this period involved the completion of the statistical analysis begun in FY 72, and collaboration on the interpretation of the results.

2. Guam Studies

A considerable amount of data processing support has been made available to members of the Epidemiology Branch in regard to surveys of morbidity and prevalence, especially in relation to the Guam studies.

Applied Neurologic Research Branch

1. Evaluation of Albutoin as an Antiepileptic Drug

Principal Investigator: Dr. J. Cereghino

The efficacy and toxicity of Albutoin was compared with diphenylhydantoin and mysoline on approximately 50 patients at New Castle State Hospital. A member of the Office of Biometry collaborated in the design and analysis of the study which was completed in a prior fiscal year. Office of Biometry support for this period involved the collaboration with Dr. Penry's staff in the preparation of a manuscript reporting the findings of the study. A member of the Office of Biometry is a co-author of the future publication.

Section on Communicative Disorders

1. Communicative Disorders Team

A member of the Office of Biometry is part of an interdisciplinary team on communicative disorders. The objective of this team is to develop a unified program plan of action in the communicative disorders area.

EXTRAMURAL PROGRAM

1. Manpower Evaluation

A member of the Office of Biometry continues to serve on the Manpower Committee of the Extramural Program of the Institute, which provides central control and guidance to the development of estimates of manpower needs in the '80's for neurologists, neurosurgeons, otolaryngologists, and speech and hearing specialists. In addition to this he also serves as a member of the Manpower Committee of the Council on Otolaryngology. In both capacities he serves as a statistician and systems analyst providing advice, and directing data processing and data reduction support. The Office of Biometry has provided forms design, systems review, and development and direction of data processing techniques in relation to the goals of the manpower project.

INTRAMURAL PROGRAM

Surgical Neurology Branch

1. Parkinson Disease - Evaluation of Therapy

Principal Investigator: Dr. J. Van Buren

Previous work on the qualitative, quantitative, and cognitive evaluations of the effect of thalamotomy on Parkinson patients was extended to the study of the usefulness of quantitative testing in the evaluation of the functional capacity of neurologically diseased patients. A sample of 20 thalamotomy-treated Parkinson patients was selected to illustrate the usefulness of quantitative (as opposed to qualitative) testing for evaluating treatment of neurological diseases. The disabilities of these 20 patients, measured as quantitative scores one month before, one week after, and annually through five years after thalamotomy were analyzed. Converted scores were analyzed by such statistical techniques as means, standard deviations, t tests, factor analyses, ANOVAS, graphs, etc.

A comprehensive review of the literature on the usefulness of quantitative testing was undertaken for use in a paper on this subject. The paper, Quantitative Testing and Its Usefulness in The Evaluation of Therapy in Neurological Diseases, has been sent to Confinia Neurologica for publication, with a member of the Office of Biometry as co-author.

2. Epilepsy Study

Principal Investigator: Dr. J. Van Buren

A study of the results of the surgical treatment of epilepsy is underway. In order to summarize the experience of the last 19 years, this project includes, (1) setting up forms for extraction and tabulation of the data from the hospital records, (2) computerizing the data for future retrieval, summary, analysis, etc.; (3) setting up an appropriate model for summarizing, analyzing, interpreting, and presenting the data; and (4) reporting the results.

3. Clinical Quantitative Testing in Neurological Studies

Principal Investigator: Dr. J. Van Buren

An investigation aimed at producing a battery of standardized clinical quantitative tests is underway. It includes examining the literature on the use of quantitative testing; examining in full detail the existing batteries of tests; selecting the mose useful portions of all existing batteries; discovering and correcting the shortcomings; standardizing the tests; formulating a revised and standardized battery of clinical quantitative tests; and formulating a proposal for using these revised and standardized tests in future studies in neurology, rheumatism, mental health and cancer.

4. Study of Nucleus Dentatus

Principal Investigator: Mr. M. Levine

Statistical support has been provided in a quantitative study of the Nucleus Dentatus (Nucleus of the Cerebellum). This entailed studying five diseased patients with longstanding diencephalic lesions and four control patients with normal cerebellum. The study included the preparation of a two-stage sampling technique for a secondary sampling of macroslide sections of the first stage sampled macroslides. Programs were prepared on the Wang 700A to obtain morphological data on the N. Dentatus, and on the time-share remote terminals to obtain additional data for comparing patients with controls to graph, analyze, summarize, and interpret the data, and to report the results for publication. Statistical techniques such as two-stage sampling techniques, random selection procedures, preparation of a special grid, a product of "square" count and "square" size for obtaining linear and volumetric measurements, t tests, ANOVAS, graphs, tables, etc., were employed to determine morphological data, neuronal count, neuronal area, etc.

5. Brain Tumor Study-Chemotherapy

Principal Investigator: Dr. A. Ommaya

Statistical analyses were performed on data from a brain tumor chemotherapy study. The study consisted of ten patients having surgery plus oral and intratumoral chemotherapy. The ten controls had surgery plus conventional therapy. The data were analyzed by t tests, chi-square tests, ANOVAS, regression analysis, covariance and graphic analysis. Survival rates for both the patient and control groups were obtained by modified life table techniques. These rates showed significant prolongation of life to approximately 50% survival after 2+ years and 25% survival after 3+ years.

6. Brain Tumor Study-Immunotherapy and Chemotherapy.

Principal Investigator: Dr. A. Ommaya

Statistical consultation and support have been provided for the analysis, interpretation, and reporting of a controlled study on brain tumor (glioblastoma) patients. Three therapy groups are to be studied with respect to the

prolongation of the life of patients with brain tumors.

In order to assess the effectiveness of the therapies as rapidly as possible and with a minimum number of patients, a sequential analysis technique was formulated. Patients will be assigned randomly to one of the three therapies and will be compared on the basis of survival time. Survival times for all groups will be compared to the already established controls (NCI), those having conventional therapy of surgery and radiation. If an EMI Scanner is obtained, data on x-ray absorption analysis of the brain, techniques for 'in vivo' analysis of brain metabolism, measurements of 'in vivo' brain tumor, etc., can be made available both for a more comprehensive study of brain tumor etiology and in conjunction with the therapy studies. Thus far, the procedure for random selection of the patients and both the graphic and tabular procedures of the sequential analyses have been presented to the investigator.

Medical Neurology Branch

1. Duchenne Muscular Dystrophy

Principal Investigator: E. Derrer

The Office of Biometry participated in the development of a screening test for drugs for treating Duchenne muscular dystrophy. The first step is to establish standard curves for several sero-enzyme responses induced by injection of serotonin in rats. Possible experimental plans have been discussed with the investigator to eliminate disturbance in the experiment. The number of animals available in each experiment group was checked for sufficiency. The implementation of this project depends on procurement of a contract laboratory to do the serological work.

2. Analysis of Graded Response Data

Principal Investigator: E. Derrer

Statistical consultation involved analysis of graded response data when the same responses were repeatedly observed in the experiment. Under this condition, the conventional methods such as t test or ANOVA may be inappropriate for testing the treatment effects. An alternative method employing contingency table approach and chi-square test was suggested.

Central Nervous System Studies Branch

1. Factors Associated with Fetal Birth and Mortality in the Rhesus Monkey, Similarities in the Human

Principal Investigator: Dr. R. DiGiacomo

In response to an article recently published in the American Journal of Obstetrics and Gynecology, which was co-authored by a member of the Office of Biometry, the investigators received data on approximately 3,000 Rhesus pregnancies, covering a period of several years. These, along with data from

the Perinatal Physiology Laboratory, form the basis for a second article currently being drafted on prematurity in the Rhesus monkey. Statistical techniques involve multivariate regression, covariance analysis, and various nonparametric procedures to test for underlying assumptions.

2. Fetal Growth Patterns in the Human

Principal Investigator: Dr. R. DiGiacomo

The investigators are working with a New Jersey obstetrical group in order to construct standard profiles (under several different sets of circumstances) for fetal growth patterns throughout pregnancy. The sampling design has been constructed, data collection instruments have been developed, and pretesting is now in its final stages. Preliminary results are expected to be available in about a year.

Neuropathology and Neuroanatomical Sciences Branch

1. Muscular Contraction Study

Principal Investigator: Dr. D. Riley

Statistical support has been provided in a muscular contraction study involving determination of the changes in several muscular contraction parameters due to electrical stimulation on muscle fibers. Muscle samples taken from different cat tails were subject to "isolation" treatment prior to stimulation. They were then stimulated electrically at frequencies of 10 HZ and 50 HZ. Muscular contraction parameters such as isometric contraction time, half relaxation time and tetanus/twitch ratio were observed on each sample and were analyzed statistically. Because of the nature of the basic variation in the experimental data, a modified t test was derived to test the significance of the effect due to stimulation. Other statistical techniques such as ANOVA of a hierarchical structure with unequal number of observations per "stimulation" group were employed to estimate variance components. A special computer program was prepared for this type of analysis. A Wilcoxon rank sum test was used to test the tetanus/twitch ratio data. A report summarizing the analytic results has been sent to the investigators for reference

2. Kinetic Analysis of Glycogen Synthesis in the Brain

Principal Investigator: Dr. J. Passoneau

The Office of Biometry participated in a study of the nature of glycogen synthesis and degradation in the brain. The turnover of brain glycogen in normal mice has been compared to that in animals treated with phenobarbital or hydrocotisone. The rate of incorporation of ^{14}C-glucose into cerebral glycogen and the subsequent loss of label has been compared with that of limit dextrin. Statistical support included: (1) a two-compartment model was developed to describe the kinetic system of accumulation and breakdown of glycogen in the brain after the validity of other alternative models has been carefully studied. The mathematical equations of the system were then

derived to determine various rate constants of incorporation and elimination of the labeled substance in the model. (2) An iterative data-fitting procedure was derived to fit the non-linear equations to the experimental data and a PDP-10 computer program (MLAB) was selected for the analysis. (3) Statistical analyses of four experiment groups were carried out (phenobarbital and hydrocortisone treated and two controls) to determine turnover rates and times for total glycogen and limit dextrin, etc.

The appropriateness and the interpretation of the proposed kinetic model was discussed with the investigators, including the experimental design for the future study. The final report "Factors Affecting the Turnover of Cerebral Glycogen and Limit Dextrin in Vivo" was reviewed prior to submission for publication.

3. Relation Between Fenistrae (holes) and Junctions in Blood Vessels From Hamster Tumors

Principal Investigator: Dr. M. Brightman

Data were collected on the number of fenistrae and the number of junctions in 42 different blood vessels from hamster tumors. The statistic of interest was the ratio of the number of fenistrae to the number of junctions. Assistance was provided in selecting the appropriate statistical estimator for this ratio as well as the standard error associated with the estimator. Since the sample was from a finite population whose size was unknown, the ordinary formula for estimating the standard error of the ratio of two means could not be used. An alternate method was provided and its rationale was explained to the primary investigator. In addition, a computer program was written and the necessary computations were carried out at the Office of Biometry.

Perinatal Physiology Branch

1. Determination of Vmax as an Index of Heart Contractile State

Principal Investigators: Drs. D. Mirvis and G. Kopf

This study is designed to determine the velocity curve for the heart contractile element (Vmax) and to extrapolate this curve back to zero pressure to get a measure of the maximum no load velocity.

The experiments have been completed and consultation is being provided to determine what methods of computing contractile indices best represent the theoretical results that should be obtained under the experimental constraints. Different insults have known different effects on contractile indices. Various graphics have been produced to aid in this analysis; work is still continuing.

2. Physiological Effects of Low Blood Pressure

Principal Investigator: Dr. D. Selkoe

The Office of Biometry provided statistical consultation and assistance

in a study dealing with the physiological behavior of rhesus monkeys during periods of low blood pressure. Interest was centered on various summary statistics which were provided after the data were computerized. Preliminary questions have been answered and results presented to the investigator.

3. The Effect of Hypoxia on the Brain in the Rhesus Monkey

Principal Investigator: Dr. H. Shapiro

A study describing some of the physiological and metabolic changes associated with a period of subtotal oxygen insult in the Rhesus monkey is underway. The key problem is the response of the brain to hypoxia.

Statistical analyses of these data are continuing; baselines for the variables have been established. Impedance measurements are now the main area of study. This study will measure the relationships between blood gas, acid-base chemistry, and the pathological performance of the insulted animals.

4. Grooming Patterns and Dominance Patterns and their Interrelationships

Principal Investigator: Dr. E. Missakian

Dr. E. Missakian and members of the Office of Biometry investigated the relationship between the grooming behavior and dominance patterns present in a free ranging colony of Rhesus monkeys.

5. Mating Patterns of Rhesus Monkeys

Principal Investigator: Dr. M. Varley

Statistical assistance was provided in a study to assess the effect of climate on the mating season of Rhesus monkey colonies on various Caribbean Islands.

CNS Studies Branch

1. Hematological and Serum Chemical Statistics for the Clinically Normal Chimpanzee

Principal Investigator: Dr. R. DiGiacomo

A study concerned with the development of normative statistics for hematological and serum chemical measurements from chimpanzees is underway. The use of statistics is two-fold:

(1) To assist in diagnosing the presence of illness in an animal in the colony.

(2) To provide a basis for testing newly-introduced animals for their "normalcy" as far as hematological and serum chemical measures are concerned.

Work has been completed and is in the hands of the principal investigator who is assessing the possibility of publication.

OTHER ORGANIZATIONS

Washington Clinic

1. Screening Method of Internal Carotid Occulsive Disease

Principal Investigator: Dr. W. Howell

The Office of Biometry participated in a project to develop a screening method for detecting internal carotid occlusive disease. The study employed a photo-plethysmography technique to record the change in volume of blood flow in both supraorbital regions as indicators of the status of occlusion. The screening procedure involves the application of thumb compression on one side of the internal carotid arteries to block the blood flowing from this side to the supraorbital regions. The recorded information thus provides a clear basis for detecting any obstruction in the non-compressed side. Seventy-five patients, whose carotid occlusive disease status has been identified by angiogram, have been recorded with photo-plethysmography. A comprehensive statistical analysis of the recorded information has been carried out which involves (1) quantifying the plethysmography recording for all patients in the study, (2) computing, for each patient, intermediate statistical measures to be used in the construction of a test criterion, (3) developing an objective test criterion through use of discriminant analysis methods, (4) examination of the efficiency of a test criterion suggested by the major investigator and of the statistically derived criterion, and (5) graphic and tabulated presentation of study results. The statistically derived test criterion is able to identify a notably higher percentage of positive patients than Howell's criterion.

A detailed study report has been prepared and submitted to Dr. MacNichol and Dr. Howell. Data from thirteen more patients have been recorded and will be further analyzed statistically to verify reproducibility of testing efficiency. The final report intended for publication will be prepared after completion of the statistical work.

RESEARCH

1. Feasibility Study to Investigate a Research Into the Problem of Missing Values

All major CPP studies encounter the problem of statistical inference using incomplete data sets. Conventional analyses applied to incomplete data may be inadequate. Several members of the Office of Biometry have examined the feasibility of a research effort directed toward the resolution of some of the issues involved. They have outlined methods for a) description of the missing data and their relationships to the variables under study, b) simulation of the missing data analysis, c) derivation of some analytical expressions for errors in estimates resulting from missing data.

2. Analysis of Hypercube Designs

Research is proceding in the analysis of variance of hypercube designs.

The research concentrates on determining which interactions in a factorial experiment with hypercube design are estimable, and on a method for examining heterogeniety and non-additivity in the experimental error variance. A literature search showed that there have been few publications in this area. Preliminary investigation has been carried out on Latin cube design, and will be extended to higher cubes.

3. Detection of Outliers

A member of the Office of Biometry is continuing research on the problem of detection of outliers. A paper on this subject will be published in the Journal of the American Statistical Association. The focus of the current work is in two areas, first, to examine the power function of the test for outliers in general linear regression and second, to extend the results to the detection of multivariate outliers.

4. The Analysis of Variance with Unequal Cell Size

A member of the Office of Biometry is doing research on the analysis of variance for the case of unequal cell sizes. This type of problem arises frequently in the analysis of biomedical data. The current methodology on non-orthogonal data is confusing. An analysis is sought that is free from the confusing attributes of the standard techniques.

MISCELLANEOUS ACTIVITIES

A. Computer and Related Items

1. Review of Time-Sharing Computer Company Proposals

Feedback from time-sharing users with NIH was sought by the Procurement Branch of DCRT in the matter of soliciting bids for commercial time-sharing computer service. Our suggestions and advice were taken into consideration in the writing of the RFP particularly in the weighting of various criteria.

RFP's were sent out, proposals were received, and the Office of Biometry is now reviewing the proposals of the five companies which use IBM 360 or 370 hardware. The decisions that result from this review are important to NIH, and particularly to the Office of Biometry since it is a heavy user of commercial time-sharing services.

2. Contract Information File

The Office of Biometry has constructed an NINDS Contract Information File and Report (CIFER) for use by the Administrative Officer of C&FR, and has developed the basis for a fully-developed information system for contract information.

3. Spike Train Analysis

The computer package SASE IV for analysis of series of events has

been studied for use in Spike Train Analysis. The Office of Biometry placed the package on the NIH computer system, and ran it with sample data. The package appears to be suitable for an initial analysis of micro-electrode recording (Spike Train) data provided by Dr. Li. The programs will be applied to these data as time permits.

4. Evaluation of Cathode Ray Tube Terminals

The Office of Biometry has studied the capabilities of cathode ray tube (CRT) high speed display and graphic terminals which might be of considerable value in the analysis of CPP and other data. The survey examined the characteristics of several of these machines and the information will be presented to statisticians and systems analysts of the Office of Biometry for comparisons and selection of a model for trial. System support for CRT display and graphics has also been investigated.

5. Computer Graphics

The Office of Biometry has provided the Extramural Program with CalComp Computer graphics which were used for slides in presentations to the NANDS Council. Similar support was provided to the C&FR Program.

6. Software Development

The Office of Biometry has maintained and furthered the development of software package systems designed to check the quality of data in a system, to create complex cross-tabulations, to build mathematical computer programs from standard sub-units, and to simplify the use of FORTRAN programming language. These software programs include Data Check, Express IV, IMSL, and Flowchart Package systems.

7. Remote Terminal Support

The Office of Biometry has provided remote terminals and programming support to expedite the various services to NINDS. It has served as a central clearance and procurement area for obtaining terminals, their maintenance, and for operations control. In addition, a remote job entry system (IBM 360/20) to the main NIH computers has been maintained as well as various auxiliary machines, such as key punches, sorters, etc. This is of direct benefit to programmers as well as to statisticians and other professionals who wish to provide some part of their own batch-data processing effort.

The Office of Biometry has also tested the Optical Scanning System (OCR) and the Computerized Microfiche Output System (COM) on the IBM 370/165 at DCRT and made them available for use.

B. Committee Activities

The Office of Biometry was represented on a number of ad hoc committees for contract review:

1. Radiologic Health Study Section of the Food and Drug Administration - a contract proposal to study the effects, on the progeny, of x-rays of the mother when the latter was herself in utero.

2. NIGMS - to site visit and review the contract proposal of the Boston Collaborative Drug Surveillance Program.

3. NICHD - a contract proposal for studies of the human menstrual cycle.

4. NICHD - contract proposals on the epidemiology of birthweight with regard to the effect of induced abortion on subsequent offspring, and on a study to substantiate the existence of hypothalamic post pregnancy syndrome.

PUBLICATIONS

Asher, D.M., Gibbs, C.J., David E. Alpers, M.P., and Gajdusek, D. C. with statistical analyses by Sadowsky, D. A.: Experimental Kuru in the Chimpanzee, Physical Findings and Clinical Laboratory Studies. Advances in Primatology, in press.

Ellenberg, Jonas H.,: The Joint Distribution of the Standardized Least Squares Residuals from a General Linear Regression. Accepted by the Journal of the American Statistical Association.

Henderson, W. G., Sadowsky, D.A., Shapiro, D.Y. and Van Buren, J.M.: Quantitative Testing and Its Usefulness in the Evaluation of Therapy in Neurological Diseases. Confinia Neurologica, in press.

London, W. T., Nahmisa, A.J., Naib, Z.M., Fuccillo, D.A., Ellenberg, J.H. and Sever, J.L.: A Non-Human Primate Model for the Study of the Cervical Oncogenic Potential of Herpes Simplex Virus Type 2, accepted by the Journal of the National Cancer Institute.

Van Buren, J.M., Shapiro, D.Y., Henderson, W.G., and Sadowsky, D.A.: A Qualitative and Quantitative Evaluation of Parkinsonism Three to Six Years Following Thalamotomy. Confinia Neurologica, 1973.

FUTURE PLANS

1. The Office of Biometry will continue its service function to NINDS in its areas of responsibility.

2. The Office of Biometry will develop a proposal which will outline the major elements of a contract program for developing precise estimates of the incidence and prevalence of diseases within the areas of NINDS responsibility.

3. The Office of Biometry will continue to study various methodological problems as part of its NINDS studies. Among the problem areas to be covered are missing data, efficient methods for dealing with large data bases and many factors, and analysis of non-orthogonal data.

ANNUAL REPORT
For Period July 1, 1972 through June 30, 1973
Perinatal Research Branch
Collaborative and Field Research
National Institute of Neurological
Diseases and Stroke
National Institutes of Health

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ANNUAL REPORT
For Period July 1, 1972 through June 30, 1973
Perinatal Research Branch
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GENERAL SUMMARY

I. INTRODUCTION:

The Collaborative Perinatal Project will complete in FY 1973 the longitudinal follow-up examinations on children at seven years of age born to 50,000 women at 12 collaborating major medical centers in the United States. The eight-year examinations will be completed in FY 1974. The basic objective of this project is to relate complications, conditions, and events that occur during a woman's pregnancy or during her labor and delivery to subsequent abnormality exhibited by the child of the pregnancy as the child grows and develops. As data collection is approaching completion, the major emphasis of the project is shifting to data analysis and interpretation for publication of reports on the Collaborative Perinatal Project research findings. In order to accomplish this, a highly coordinated effort is being planned to meet the basic data analysis objectives of the Collaborative Perinatal Project. To provide a framework for this effort, a comprehensive plan for data analysis and interpretation has been written.

II. DATA COLLECTION:

The Collaborative Perinatal Project was initiated in 1959 when the women began registering at each of the collaborating institutions during their pregnancies. Women continued to enroll in the study through December of 1965. The babies were delivered between 1959 and 1966 and received several examinations during their first year of life, at 3 years of age, and at 4 years of age. The 7-year extensive battery of examinations includes a pediatric-neurological examination; a battery of psychological tests, including an IQ determination; and a visual screening test. These will have been completed on all study children by the end of FY 1973. An additional examination at eight years of age, an assessment of speech, language and hearing development, will be completed on children at five of the collaborating institutions by June of 1974. With data collection soon to be complete, the basic analysis pertinent to the objectives of the study is being implemented.

III. A COMPREHENSIVE PLAN FOR ANALYSIS AND INTERPRETATION OF COLLABORATIVE PERINATAL PROJECT DATA:

Broadly stated, the Collaborative Perinatal Project is concerned with the identification of prenatal factors that have a sufficiently high association with adverse pregnancy outcome and subsequent neurological and mental development of the child to provide leads to the etiologies

of the abnormalities and thus to the development of strategies for prevention and intervention. After a careful review of the objectives of the Collaborative Perinatal Project, the data available for analysis and the work in progress, it was recommended that major efforts in analysis and interpretation are needed in ten primary areas in order to meet the basic objectives of the project. Monograph reports in book form are planned in each of the following areas: Cerebral Palsy; Mental Retardation; Communicative Disorders; Visual Abnormality; Convulsive Disorders; Learning Disorders; Minimal Brain Dysfunction; Congenital Malformations; Birthweight-Gestational Age Relationships (Prematurity); and Neuropathology, General Pathology and Placentology.

IV. IMPLEMENTATION:

Implementation of the Comprehensive Plan for Analysis and Interpretation of Collaborative Perinatal Project Data will be through teams of researchers, headed in each of the ten areas by a member of the Professional Staff Consultants Section of the Perinatal Research Branch. On each team will be a member of the Office of Biometry staff, who will participate fully in the development of the analysis. In addition, a member of the Section for Production of Data Analysis, Perinatal Research Branch, will be assigned to each primary area to facilitate data processing. The assignments are as follows:

PRIMARY DATA ANALYSIS AREA	PERINATAL RESEARCH BRANCH	OFFICE OF BIOMETRY
Cerebral Palsy	Dr. K. B. Nelson	Dr. J. H. Ellenberg
Mental Retardation	Dr. S. H. Broman	Dr. P. W. Shaughnessy
Communicative Disorders	Dr. P. J. LaBenz	Mr. D. Rubinstein
Visual Abnormality	Dr. R. Feinberg	Dr. B. F. McDonagh
Convulsive Disorders	Dr. K. B. Nelson	Dr. B. F. McDonagh
Learning Disorders	Dr. S. H. Broman	Dr. P. W. Shaughnessy
Minimal Brain Dysfunction	Dr. P. L. Nichols	Dr. T. C. Chen
Congenital Malformations	Dr.N.C.Myrianthopoulos	Dr. J. H. Ellenberg
Birthweight-Gestational Age Relationships (Prematurity)	Dr. B. H. Williams	Miss E. C. Jackson
Neuropathology, General Pathology and Placentology	Dr. J. S. Drage	Dr. T. C. Chen

In each of the ten primary areas, a program plan is being developed which will expand in detail on the summary statements in the comprehensive plan and will give a detailed approach to the analysis and identify major components. Milestone charts will be designed to set goals and to aid in the measurement of progress. Written monthly reports will be prepared to record progress and identify problem areas.

In order to facilitate the primary data analyses and/or to complete major efforts well underway, analyses are to be completed in the following secondary areas: Toxemia; Maternal Infection During Pregnancy; Labor and Delivery; Neonatal Hyperbilirubinemia; Maternal Anesthesia-Analgesia During Labor and Delivery; Four-Year IQ (to be published in 1973); Physical Growth and Development (Birth to Seven Years); Twins; Genetic and Socioeconomic Factors; and Drugs Taken During Pregnancy.

V. WORK IN PROGRESS:

One of the major activities within the Perinatal Research Branch during FY 1973 has been a study of the relationship between intelligence at four years of age, as measured by the Stanford-Binet Intelligence Scale, and a variety of factors related to pregnancy, delivery, postnatal status, and environment. A substantial monograph has been developed and has received preliminary review by outside reviewers. The manuscript is currently undergoing revision, and when this is complete, NINDS clearance will be sought.

A study of the genetics of intellectual and motor performance has been partially reported in a number of publications, and several other papers are in preparation. The objective of these studies is to assess the contribution of genetics to the variance of behavioral measures, particularly intellectual and motor performance, by correlating scores and measurements of twin, sibling, and half-sibling pairs.

Bio-social factors, associated with communicative disorders and competence in children are being studied on data collected on about 15,000 children at age 3 and additional data are being collected at age 8. Preliminary tabulations of 3-year-old data have been made by examination items and by combinations of such items as scored according to protocol criteria and according to revised criteria. Correlation matrices were constructed to explore the relationship among three-year-old subtest scores. Pilot frequency distributions were run on a small sample of 8-year-old tests prior to large sample tabulations. The development of three indices is being pursued.

Physical growth and development is being studied by the development of normative distributions, ratios and correlated coefficients of certain physical growth characteristics of children from birth to seven years. Birthweight-gestational age characteristics are being reviewed, specifically in regard to small-for-date and large-for-date babies. Work continues on the bivariant analysis of growth measurements.

Work has progressed on the comprehensive analysis of congenital malformations. These studies deal with the epidemiology of congenital malformations in singletons and twins, a study of the effect of medical, genetic and socioeconomic factors on the occurrence of congenital malformations, and an analysis of the effects of maternal diabetes on the occurrence of congenital malformations in the offspring.

A unique file has been prepared by reviewing records of women who reported relatives in the Collaborative Perinatal Project, creating a file of family-linkage groups. Over 3,000 of such cases exist, and this file will be essential to the analysis of genetic and congenital malformations.

All children identified by the age of one year who had a definite convulsion were studied. Perinatal data, subsequent seizure history, 4- and 7-year IQ's were examined and several subgroups within the heterogeneous clinical category of febrile seizures were identified. The risk of later sequelae was analyzed by subgroup, and several clinical features could be isolated which identified children with significantly higher risk for later afebrile convulsive disorders and mental impairment.

VI. CONTRACT DEVELOPMENT:

One of the major complications of pregnancy associated with an increased incidence of neonatal mortality is toxemia. The initial cross-section analysis of the data on hypertension, edema, and proteinuria and their combinations was reported in the Annual Report for FY 1972. The professional staff consultant in obstetrics resigned in August 1972, and further research effort on toxemia will be via the contract mechanism. The Perinatal Research Branch is currently evaluating competitive contract proposals for a longitudinal study of the clinical signs of toxemia in pregnancy.

The analysis of the neuropathology, general pathology and placentology collected within the framework of the Collaborative Perinatal Project will be shifted from the Section on Pathology to contractors. In view of this decision, the Section on Pathology will terminate as of June 1, 1973, by which time it is anticipated that the contracts for analysis of the pathology material will be operational.

VII. OTHER FUNCTIONS:

The Perinatal Research Branch continues to monitor sample maintenance, quality control, overdue forms, data processing and institutional data collection contracts.

CONTRACT NARRATIVE
Perinatal Research Branch -- Professional Staff Consultants Section
July 1, 1972 through June 30, 1973

UNIVERSITY OF MICHIGAN (NIH-NINDS 71-2390)

Title: Genetic typing on children in NIH Perinatal Project

Contractor's Project Director: H.G. Gershowitz, Ph.D.

Current Annual Level: None

Objectives: To obtain approximately 5,000 blood specimens from children born in the Collaborative Study, separate red cells from serum and freeze both in liquid nitrogen for further studies.

Major Findings: None

Current Status of the Contract: The contract has been extended without funds for one year. Only about 600 blood specimens have been collected. Because of the difficulty of obtaining blood specimens the contract will not be continued. No decision has been made yet as to the disposition of the blood specimens and the equipment.

CONTRACT NARRATIVE
Perinatal Research Branch -- Professional Staff Consultants Section
July 1, 1972 through June 30, 1973

UNIVERSITY OF WISCONSIN (NINDS-NIH 72-2303)

Title: Genetic-odontometric study of pre and neonatal growth

Contractor's Project Director: Richard Osborne, Ph.D.

Current Annual Level: $186,095.00

Objectives: To correlate the incidence and prevalence of dental and facial abnormalities with neurological defects, congenital abnormalities and other disorders of childhood.

Methodology: Dental casts and photographs of teeth will be obtained on about 4000 Collaborative Study children who are scheduled for routine seven or eight year examinations at selected participating institutions, and on children rescheduled because of known complications during their prenatal and neonatal periods of study.

Major Findings: During the first year of study about 22 percent of children were found to have dental abnormalities.

Significance to Program: The study of dental abnormalities is expected to yield an index of stress or abnormality during prenatal development.

Proposed Course of Contract: During the first year a little over 1000 casts and photographs have been taken. These were sent to the University of Wisconsin where they were examined for quality and for defects, catalogued and stored. During the second year collection of the material will be completed and plans will be made for analysis.

CONTRACT NARRATIVE
Perinatal Research Branch, C&FR, NINDS
Office of the Chief
July 1, 1972 through June 30, 1973

UNIVERSITY OF MICHIGAN (NIH-NINDS-72-2320)

Title: Bivariant Physical Growth

Contractor's Project Director: Stanley M. Garn, M.D.

Current Annual Level: $16,900

Objectives:

1. Analyze the physical measurement data by preparing tables on children from the Collaborative Perinatal Project.

2. Develop the following types of tables:

   a. Sex and race-specific means, standard deviations and percentiles by age, for (1) weight, (2) total body length, (3) head circumference, (4) chest circumference.

   b. Sex, race and age-specific tables for (1) weight percentiles, (2) head circumference percentiles and (3) chest circumference percentiles versus specified height intervals.

   c. Sex, race and age-specific tables for head circumference percentiles versus weight intervals as specified.

   d. Age, sex and race-specific tables for mean weights for a matrix of 5 chest sizes by 24 total body lengths for groups of sufficient sample size.

Course of Contract: June 28, 1972 through September 27, 1973.

CONTRACT NARRATIVE
Perinatal Research Branch, C&FR, NINDS
Office of the Chief
July 1, 1972 through June 30, 1973

JOHNS HOPKINS UNIVERSITY (NIH-NINDS-72-2321)

Title: Ancillary Studies

Contractor's Project Director: Janet B. Hardy, M.D.

Current Annual Level: $49,993

Objectives:

Using NINDS Collaborative Perinatal Project data and data unique to the Johns Hopkins Collaborative Perinatal Study, the contract will conduct studies in the following areas:

a. Perinatal infections.
b. Problems in communication.
c. Growth and development of the child in an inner-city population.
d. The psychological, biological and environmental factors relating to I.Q.
e. The etiology and characterization of seizures.
f. The effect of maternal smoking on the fetus.
g. The toxicity of low levels of bilirubin.

Course of Contract: June 28, 1972 through June 27, 1975.

CONTRACT NARRATIVE
Perinatal Research Branch, C&FR, NINDS
Office of the Chief
July 1, 1972 through June 30, 1973

BOSTON UNIVERSITY (NIH-NINDS-72-2322)

Title: Maternal Drug Ingestion and Fetal Abnormalities

Contractor's Project Director: Dennis Slone, M.D.

Current Annual Level: $149,000

Objectives:

Using NINDS Collaborative Perinatal Project data, the objective of the contract is to conduct a research program on maternal drug ingestion and fetal abnormalities. The study will describe maternal drug utilization patterns, provide for a rapid evaluation of drug teratogenicity, report on the relationship between Dilantin and selected congenital malformations, investigate the possible teratogenic effects of virus vaccines, and investigate possible associations between drugs and congenital cardiac malformations.

Course of Contract: June 28, 1972 through June 27, 1975.

Serial No. NDS (CF)-63 PR/OC 1144
1. Perinatal Research Branch
2. Office of the Chief
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: An Instrument For The Conduct of A Retrospective Study of Seizures, Cerebral Palsy, Mental Retardation and Other Neurological and Sensory Disorders of Infancy and Childhood.

During FY '73 this project was transferred to Office of Director, NINDS, Serial No. NDS OD/OPE 1144. The principal investigator is now situated there, but the project will continue to be reported under Perinatal Research Branch. (Please see pages 115-z - 116-z)

Serial No. NDS (CF)-63 PR/OC 1146
1. Perinatal Research Branch
2. Office of the Chief
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Public Health Implications Study of Perinatal Mortality in the Collaborative Study and in the Collaborative Study Cities.

During FY'73 this project was transferred to Office of Director, NINDS, Serial No. NDS OD/OPE 1146. The principal investigator is now situated there but the project will continue to be reported under Perinatal Research Branch. (Please see pages 117-z-118-z)

Serial No. NDS (CF)-73 PR/OC 2052
1. Perinatal Research Branch
2. Office of the Chief
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: The First Year of Life

Previous Serial Number: None

Principal Investigator: Joseph S. Drage, M.D., PRB, C&FR, NINDS

Other Investigators: Janet B. Hardy, M.D., Associate Professor of Pediatrics, The Johns Hopkins University
Esther Jackson, Office of Biometry, C&FR, NINDS

Cooperating Units: Johns Hopkins University; Office of Biometry, C&FR, NINDS

Man Years:

Total:	.02
Professional:	.01
Other:	.01

Project Description:

Objectives: "The First Year of Life" is planned as a volume to report on the frequency distribution of a number of findings reported on Collaborative Perinatal Project children during the first year of their lives. It will include information on birthweight-gestation distributions, bilirubin levels, age at hospital discharge, and distributions of various pathological findings detected during the nursery stay and during the first year of life. Of particular interest will be information regarding brain abnormality as detected during the nursery period. This volume is intended to serve as a general description of the Collaborative Perinatal Project children during their first year of life and as a reference document for further in-depth studies.

Honors and Awards: None

Publications: None

Serial No. NDS (CF)-63 PR/PSC 1163
1. Perinatal Research Branch
2. Professional Staff Consultants Section
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: An Investigation into the Relationship Between Congenital Heart and Great Vessel Anomalies and Selected Factors as Recorded in the Collaborative Perinatal Research Project.

Previous Serial Number: Same

Principal Investigator: Lenore Bajda, M.D., PRB, NINDS

Other Investigators: None

Cooperating Units: Office of the Director, NHI

Man Years:

Total:	0
Professional:	0
Other:	0

Project Description: The primary objective of this study is to assess relationships between certain maternal variables and congenital heart-great vessel anomalies.

Additional objectives include investigating relationships between early signs of abnormality and the existence of definitive congenital heart lesions and determining the existence of congenital heart-vessel anomaly in conjunction with mental retardation as recorded in the eight-month psychological examination, the one-year summary records, four-year psychological examination, and seven-year neurological and psychological examinations.

To date, maternal parameters analyzed included age of gravida, parity, prior pregnancy outcome, prior and current health status, ABO blood group, current smoking pattern, and viral antibody status.

Data was used from Collaborative Study records received by PRB from the onset of the Study (January 1959) through December 1964. These records provided 112 live and stillbirth cardiac cases for study out of a population pool of approximately 38,000. Preliminary analysis indicated that there was a definite preponderance of mothers over 30 in the C-V Study group. Controlling for race, and removing cases with chromosomal aberrations, there were more white mothers in the 30 and over age group than expected at the .05 level. This trend is also noted among Negroes. There was a greater than expected number of gravida

with systemic disease complications and prior pregnancy loss among the mothers of the cardiacs. A breakdown of these factors for greater specificity is pending.

Because the number of patients with each specific cardiac abnormality was small, specific associations between serological findings and clinical observations were not possible, although several interesting trends were noted.

A preliminary report on the 1964 cohort study was presented at the 1965 APHA Annual Meeting and the 1966 Annual Meeting of the Teratology Society.

Analysis of an expanded Study cohort through 1967, with a population pool of approximately 55,000 providing additional cases, is underway in anticipation that the additional cases will support earlier findings and perhaps provide further clues for identifying etiological relationships.

The urgent priority of processing the entire Collaborative Project study data has temporarily delayed further work on this study.

Honors and Awards: None

Publications: None

Serial No. NDS (CF)-63 PR/PSC 1184
1. Perinatal Research Branch
2. Professional Staff Consultants Section
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Population Dynamics of Tay-Sachs Disease and other Sphingolipidoses

Previous Serial Number: Same

Principal Investigator: Dr. N.C. Myrianthopoulos, PRB, NINDS

Other Investigators: Dr. Stanley Aronson, Brown University

Cooperating Units: None

Man Years:

Total:	.05
Professional:	.05
Other:	.00

Project Description:

Objectives: To confirm experimentally the epidemiologic finding that the selective advantage of the Jewish TSD heterozygote is due to possible protection of the heterozygote from tuberculosis.

Methodology: It is planned to measure the rate of growth of the mycobacterium tuberculosis in media with and without hexosaminidase A.

Major findings: None

Honors and Awards: None

Publications: Myrianthopoulos, N.C., Naylor, A.F., and Aronson, S.M.: Founder effect in Tay-Sachs disease unlikely. J. Hum. Genet. 24:341-342, 1972.

Myrianthopoulos, N.C. and Aronson, S.M.: Population dynamics of Tay-Sachs disease. II. What confers the selective advantage upon the Jewish heterozygote? In Volk, B.W. and Aronson, S.M. (Eds.) Sphingolipids, Sphingolipidoses and Allied Disorders, Plenum, New York, 561-570, 1972.

Serial No. NDS (CF)-65 PR/PSC 1274
1. Perinatal Research Branch
2. Professional Staff Consultants Section
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Genetic Bases of Neonatal Reflexes

Previous Serial Number: Same

Principal Investigator: Dr. A.F. Naylor, PRB, NINDS

Other Investigators: Dr. N.C. Myrianthopoulos, PRB, NINDS

Cooperating Units: None

Man Years:

Total:	.01
Professional:	.01
Other:	.00

Project Description:

Objectives: To investigate the validity of regarding the suck, rooting and other neonatal reflexes as genetic entities.

Major findings: An initial set of cases retrieved for absence of one or more of these reflexes was reviewed and seemed to have high frequencies of various kinds of trauma whose base line frequencies were unknown.

Proposed course: To place limits on the frequencies of losses of suck, rooting, palmar grasp, plantar grasp and Moro reflexes because of mutation or segregation at gene loci specifically affecting manifestation of these reflexes.

The completion of the (condensed) Variable Data File makes practical the reactivation of this project along proper lines. Base populations can be selected for general health, especially neurological, and frequencies of isolated absence or weakness of single neurological signs can be tested. Active work on this project will be undertaken when most current tasks have been carried out.

Honors and Awards: None

Publications: None

Serial No. NDS (CF)-65 PR/PSC 1276
1. Perinatal Research Branch
2. Professional Staff Consultants Section
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Sequential Aspects of Occurrence of Spontaneous Abortion in Family Histories

Previous Serial Number: Same

Principal Investigator: Dr. A.F. Naylor, PRB, NINDS

Other Investigators: Dr. Dorothy Warburton, College of Physicians and Surgeons of Columbia University

Cooperating Units: None

Man Years:

Total:	.15
Professional:	.15
Other:	.00

Project Description:

Objectives: To relate the risk of spontaneous abortion to maternal age and prior reproductive experience. A special point under investigation is whether apparent age effects are explicable by a tendency for intrinsic habitual aborters to remain in the reproductive population longer in attempts to compensate for unsuccessful pregnancies. Also conditional risks have been estimated.

Proposed course: Although certain portions of the data analysis support a maternal aging explanation for part of the trend in abortion risk, a reconsideration of the original tabular outputs shows that a very substantial additional parity exists. An existing manuscript has been completely redrafted to reflect this change in interpretation and has been submitted for publication.

Honors and Awards: None

Publications: None

Serial No. NDS (CF)-66 PR/PSC 1338
1. Perinatal Research Branch
2. Professional Staff Consultants Section
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: The Association of Mental Subnormality with Head Circumference, Congenital Malformations, and Other Conditions of the Newborn Term Infant

Previous Serial Number: Same

Principal Investigator: Lenore Bajda, M.D., PRB, NINDS

Other Investigators: None

Cooperating Units: Office of Biometry

Man Years:

Total:	0
Professional:	0
Other:	0

Project Description: The objective of this study is to determine the relationship between head size and certain other physical features of the Collaborative Study child noted shortly after birth, at the one-year examination and at 4 years upon completion of the psychological examination. The project was in abeyance pending updating of the data file. Dr. Nelson and Mr. Deutschberger completed their study on "Head Size at One Year as a Predictor of Four-Year IQ" (Develop. Med. Child Neurol. 12: 487-495, 1970) using a sample of the Collaborative Study population including a partially selected pediatric group of 9,379 children. They concluded that there is approximately a 50% chance of an IQ of less than 80 at 4 years of age for the one-year male with a head size of less than 43 cm. and a one-year female with a head size of less than 42 cm. The investigator plans to examine in detail the low and high head measure sample of the Nelson-Deutschberger study for other factors which might account for the correlation between head size and the four-year IQ values. Depending on the results of this analysis, the original study proposal will then move forward with a larger sample, or terminate.

The urgent priority of processing the entire Collaborative Project study data has temporarily delayed further work on this study.

Honors and Awards: None

Publications: None

Serial No. NDS (CF)-67 PR/PSC 1510
1. Perinatal Research Branch
2. Professional Staff Consultants Section
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: The Study of Maternal Effects in the Production of Congenital Malformations

Previous Serial Number: Same

Principal Investigator: Dr. N.C. Myrianthopoulos, PRB, NINDS

Other Investigators: None

Cooperating Units: None

Man Years:

Total:	.05
Professional:	.05
Other:	.00

Project Description:

Objectives: To determine the extent to which maternal factors are involved in the production of congenital malformations, and to single out those malformations and conditions of the newborn in which the role of maternal factors, genetic and environmental, appears to be deciding.

Methods employed: The GEN 5-8 was used to obtain a history of outcome of prior pregnancies, in families in which half-siblings are present. Study pregnancies were also included.

Major findings: In a pilot study six conditions were found to occur in high frequency among half sibs: Rh trouble, convulsions, congenital heart disease, club foot, mental retardation and polydactyly. Analysis of the distribution of these abnormalities in first and second sibships of the same family showed that in the case of seizures and mental retardation the experience in the first sibship was significantly different from that in the second, indicating a genetic etiology; in the case of congenital heart defects and club foot the experience seemed to be the same in both sibships, indicating that environmental maternal factors predominate.

Current status: This study has now been incorporated in the comprehensive study of congenital malformations. Since some of the malformations and conditions cannot be identified until later in childhood, analysis will be resumed when the 7-year data become available.

Honors and Awards: None

Publications: Myrianthopoulos, N.C.: Report in Proceedings of Workshop of Epidemiology of Epilepsy, M. Alter Editor, in press.

Serial No. NDS (CF)-67 PR/PSC 1514
1. Perinatal Research Branch
2. Professional Staff Consultants Section
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Record Linkage of Relatives Registered in the Collaborative Study

Previous Serial Number: Same

Principal Investigator: Dr. A.F. Naylor, PRB, NINDS

Other Investigators: Dr. N.C. Myrianthopoulos, PRB, NINDS

Cooperating Units: None

Man Years:

Total:	.25
Professional:	.25
Other:	.00

Project Description:

Objectives: To identify all relatives of gravidae registered in the Collaborative Study.

Methodology: Keypunched reports of relatives have been edited for many errors and the NINDS serial numbers of the registrants have been condensed into non-redundant family groupings. Data records from a previously created version of the Variable File (Varfile) have been selected and resequenced to correspond to the family groupings. Thus, quite usable genetically grouped data can now be accessed.

Proposed course: Systematic errors in renumbering non-Core NINDS numbers and some suspiciously large family groups require review and correction before all restrictions on use of the files can be removed.

Honors and Awards: None

Publications: None

Serial No. NDS (CF)-67 PR/PSC 1515
1. Perinatal Research Branch
2. Professional Staff Consultants Section
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Rh Hemolytic Disease in Negro and White Infants

Previous Serial Number: Same

Principal Investigator: Dr. A.F. Naylor, PRB, NINDS

Other Investigators: None

Cooperating Units: None

Man Years:

Total:	.01
Professional:	.01
Other:	.00

Project Description:

Objectives: To confirm a report that high Rh antibody levels have smaller morbid effects in Negro than in white babies, although this is not true for ABO antibodies.

Major findings: Preliminary and indirect confirmation has been obtained, from a small data sample under study in this Section, for reports in the literature that high Rh antibody titers are not as highly associated with serious morbidity in Negroes as in whites.

Proposed course: The existence of the Variable Data File will make possible the easy execution of the required data processing.

Honors and Awards: None

Publications: None

Serial No. NDS (CF)-67 PR/PSC 1516
1. Perinatal Research Branch
2. Professional Staff Consultants Section
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Size of Placenta in Relation to Mother-Fetus Antigenic Difference

Previous Serial Number: Same

Principal Investigators: Dr. Dorothy Warburton, College of Physicians and Surgeons of Columbia University
Dr. A.F. Naylor, PRB, NINDS

Other Investigators: Dr. G. Nicholas Rogentine, Immunology Branch, NCI
Dr. Robert Cefalo, Obstetrics, National Naval Medical Center
Ms. Doris Mahoney, PRB, NINDS

Cooperating Units: Obstetrics Department, National Naval Medical Center

Man Years:

Total:	.50
Professional:	.20
Other:	.30

Project Description:

Objectives: To investigate the hypothesis published by Billington in 1964, that sensitization of the mother during pregnancy against paternal antigens leads to non-pathological placental hypertrophy and increased birthweight in succeeding pregnancies.

Major findings: Analysis of a large sample of study data has given convincing support to the hypothesis. A paper reporting the evidence has been published.

Proposed course: The arrangements for a laboratory study, described in the 1969-70 Annual Report have been active and 44 maternal sera have been tested against paternal leucocytes. Logistic complications which cause poor data return for effort have stopped collection of new cases. The accumulated data will soon be reviewed for publishability.

Honors and Awards: None

Publications: None

Serial No. NDS (CF)-69 PR/PSC 1748
1. Perinatal Research Branch, NINDS
2. Section on Professional Staff Consultants
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Neonatal Polycythemia: I. A Manifestation of Chronic Injury During Distress

Previous Serial Number: NDS (CF)-69 PR/PN 1748 and incorporating Serial No. NDS (CF)-69 PR/PN 1749

Principal Investigator: Miles M. Weinberger, M.D., Greenbrae, California

Other Investigators: Arthur Oleinick, M.D., Portland, Oregon
John A. Churchill, M.D., Harper Hosp., Detroit,MI

Cooperating Units: None

Man Years:

Total:	0
Professional:	0
Other:	0

Project Description: The objectives of the study were to study demographic and maternal factors associated with neonatal polycythemia.

As part of the protocol during the Collaborative Study on Cerebral Palsy, capillary hematocrits were obtained as near to 48 hours as possible (generally between 36 and 60 hours) on 44,683 newborns, or 86% of all 77 and over were identified, and controls, matched for institution and year of birth, race, sex, socioeconomic index, and presence or absence of 4-year follow-up examination were selected randomly from all infants with 48-hour hematocrits 50 through 65. Various demographic and maternal factors were identified in both subjects and controls, and differences were statistically evaluated.

The subject cases (those with polycythemia manifested by hematocrits 77 and over) were found to have been the product of longer gestation, but were smaller in weight than the control population. There was an increase in incidence of placental pathology and the placenta of the subject cases was significantly lighter than the weights of the controls. One-minute and five-minute Apgar scores were both lower in the subject cases and there was a greater incidence of dysmaturity diagnosed in the subject cases. When compared with the whole Collaborative Study population, infants with polycythemia were noted to come from lower socioeconomic groups. Data suggest that neonatal polycythemia may be manifestation of chronic intrauterine distress.

A manuscript has been written for this study and is now in review. Due to the scattering of the authors and their other obligations, the review process has not progressed too far.

Honors and Awards: None

Publications: None

Serial No. NDS (CF)-70 PR/PSC 1850
1. Perinatal Research Branch
2. Professional Staff Consultants Section
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: The Clinical Signs of Toxemia of Pregnancy and Their Association with the Outcome of Pregnancy

Previous Serial Number: NDS (CF)-70 PR/OB 1850

Principal Investigator: This project is in the process of being converted to a contract study since we do not currently have an obstetrician on the PRB professional consultant staff.

The adequacy (consistency and validity) of the clinical data available, the range of their variablilty at seven intersections in the course of pregnancy and their association with certain population characteristics (race, age, parity) has been demonstrated.

The next step in the analysis of toxemia is to study the occurrence, increase, decrease and appearance over time of the clinical signs of toxemia in the course of individual study pregnancies and in association with the immediate and long term outcome of pregnancy.

Methods: Identification of time patterns and combinations.

Honors and Awards: None

Publications: None

Serial No. NDS (CF)-70 PR/PSC 1857
1. Perinatal Research Branch
2. Professional Staff Consultants Section
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: The Genetics of Intellectual and Motor Performance

Previous Serial Number: Same

Principal Investigator: Sarah H. Broman, Ph.D., PRB-NINDS

Other Investigators: N. C. Myrianthopoulos, Ph.D., PRB-NINDS
V. L. Anderson, Ph.D., University of Minnesota
Paul L. Nichols, Ph.D., PRB-NINDS

Cooperating Unit: The Dight Institute for Human Genetics, University of Minnesota

Man Years:

Total:	.05
Professional:	.02
Other:	.03

Project Description:

Objectives: The objective of the study is to assess the contribution of genetics to the variance of behavioral measures, particularly intellectual and motor performance, by correlating scores and measurements of twin, sibling and half-sibling pairs. The results of the psychological tests given at ages eight months, four years and seven years have been analyzed. As a physical parallel, correlations for height, weight and head circumference at several ages have been computed.

Some selected findings are as follows: (1) heritability of IQ at age four does not appear to be lower than in older populations; (2) the between family variance componant of the four-year IQ test was nearly twice as large in the white population as in the Negro population suggesting that heritability is lower in Negroes than in whites; (3) an analysis of variance of weight and height measured at four years revealed that, unlike IQ, the variance componants were not different in whites and Negroes; (4) at seven years a high correlation (.86) was found between 12 subtests social class loading and the Negro-white differences on the test, while no relationship was found between Negro-white differences and heritabilities of the subtests after controlling for the subtests' social class loading; (5) the discrepancy in IQ between twins and singletons decreases in both races

from four to seven years; (6) differences in intellectual performance between white and Negro children of similar socioeconomic backgrounds were negligible; (7) the influence of genetics on scores on the 8-month Bayley Mental Scale is most important at the low end of the distribution.

Analyses and the preparation of several papers are continuing.

Honors and Awards: None

Publications:

Nichols, P. L. The effects of heredity and environment on intelligence test performance in 4 and 7 year old white and Negro sibling pairs. Doctoral dissertation, University of Minnesota, 1970.

Myrianthopoulos, N. C., Nichols, P. L., Broman, S. H., and Anderson, V. L.: Intellectual development of a prospectively studied population of twins. In Proceedings of the Fourth International Congress of Human Genetics, Amsterdam, Excerpta Medica, in press.

Nichols, P. L. and Anderson, V. L. Intellectual performance, race and socioeconomic status. Soc. Biol., in press.

Serial No. NDS (CF)-71 PR/PSC 1904
1. Perinatal Research Branch
2. Professional Staff Consultants Section
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: The Study of Labor and Delivery

Previous Serial Number: NDS (CF)-71 PR/OB 1904

Principal Investigator: Primarily a Task Force Investigation
Dr. Vincent Tricomi, Chairman

Other Investigators: Emanuel A. Friedman, M.D., Harvard University Medical School, Boston, Mass.
Luke Gillespie, M.D., Boston Lying-in Hospital, Boston, Mass.
Marvin Green, M.D., New York Medical College, New York, New York
Esther Jackson, Office of Biometry, NINDS
Schuyler G. Kohl, M.D., State University of New York, Downstate Medical Center, Brooklyn, N.Y.
Kenneth R. Niswander, M.D., University of California at Davis, Calif.
David Rubinstein, Office of Biometry, NINDS
Vincent Tricomi, M.D., Brooklyn-Cumberland Medical Center, Brooklyn, N.Y.

Cooperating Units: All institutions participating in the Collaborative Project

Man Years

Total:	0.2
Professional:	0.1
Other:	0.1

Project Description:

This study has two objectives:

A. To determine the influence of specific labor and delivery factors on the fetus in terms of immediate outcome and later neurologic and psychologic development.

B. To develop a reference standard of actuarial, obstetric, and labor and delivery features that will result in optimal outcome.

Labor is usually reported in quantitative units of hours and minutes and is conventionally divided into short, normal, and long labor by arbitrarily selected time intervals. These intervals, in turn, have been associated with certain complications of labor. FRIEDMAN has demonstrated in a long series of publications that these associations do not necessarily reflect the underlying physiology or pathology of uterine activity. He devised a methodology to quantify uterine activity and its deviations in relation to the phases of labor, which are based upon the observed progress of labor in time. Possible damage to the fetus is not necessarily dependent upon the total duration of labor, but on the dyscoordination or changes in the duration of any one or combinations of the phases of labor.

The identification of gravidae with specified types of labor will produce several standard cohorts that may be used by other Study sections and task forces (Pediatric-Neurology, Pathology, Physical Growth and Development, etc.) as variables for their respective special studies.

Methodology:

A reference cohort of gravidae with "normal labors" is defined, for which the specified outcome will be tabulated. This reference cohort will be compared with cohorts of specified types of dysfunctional labor and their respective outcomes.

Those gravidae with incomplete data on labor and delivery in the Study record will be identified and their general characteristics (race, age, parity, sex of fetus) and outcomes will be tabulated separately.

Initial computer printouts have been obtained and are in the process of being analyzed.

Honors and Awards: None

Publications: None

Serial No. NDS (CF)-71 PR/PSC 1906
1. Perinatal Research Branch
2. Professional Staff Consultants Section
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Blood group effects in mother and offspring

Previous Serial Number: Same

Principal Investigators: Dr. N.C. Myrianthopoulos, PRB, NINDS
Dr. Bernice Cohen, Johns Hopkins University

Other Investigators: None

Cooperating Units: None

This project has been discontinued.

Serial No. NDS (CF)-71 PR/PSC 1907
1. Perinatal Research Branch
2. Professional Staff Consultants Section
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Congenital malformations in the Collaborative Study
I. Epidemiologic survey in singletons

Previous Title: Epidemiologic and genetic study of congenital malformations

Previous Serial Number: Same

Principal Investigators: Dr. N.C. Myrianthopoulos, PRB, NINDS
Dr. C.S. Chung, University of Hawaii

Other Investigators: None

Cooperating Units: None

Man Years:

Total:	.40
Professional:	.30
Other:	.10

Project Description: This project has grown in size and scope so that it became necessary to separate it into four different projects. This first part is retained as the original project; the other three are described as new projects.

Objectives: To study the epidemiology of all congenital malformations in single born children in the Collaborative Study, to determine racial and sex differences in the incidences of these malformations and to examine institutional variability.

Proposed course and methodology: A definitive list of congenital malformations and a file of all Collaborative Study children who had malformations at birth or during the first year of age have been developed. Malformations are divided into major and minor. Analyses are done by race and sex.

Major findings: About 15 percent of children born in the Collaborative Study had one or more malformations. Half of the malformations were major. Only about a third of malformations observed during the first year of life were diagnosed at birth. Except for three minor malformations which were more frequent among Negroes, there were no significant differences in malformation incidences between Negroes and whites.

Current status: This study is now completed and a manuscript is being prepared.

Honors and Awards: None

Publications: None

Serial No. NDS (CF)-71 PR/PSC 1908
1. Perinatal Research Branch
2. Professional Staff Consultants Section
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: A chromosomal study of children in the Collaborative Study

Previous Serial Number: Same

Principal Investigators: Dr. N.C. Myrianthopoulos, PRB, NINDS
Dr. H. Lubs, University of Colorado

Other Investigators: None

Cooperating Units: Boston Children's Hospital Medical Center, Children's Hospital University of Buffalo, University of Tennessee, Children's Hospital of Philadelphia and University of Oregon Medical School

Man Years:

Total:	.10
Professional:	.05
Other:	.05

Project Description:

Objectives: To study the epidemiology of chromosomal aberrations in approximately 10,000 Study children at age 7 years; and to relate major and minor chromosomal deviants to growth, mental and motor development and neurological status of these children.

Proposed course and methodology: A blood sample is taken from children at the five collaborating institutions, during their 7-year examination. Two cells are immediately analyzed; ten cells are analyzed when chromosomal anomalies are found or when the children have some mental or motor anomaly or a congenital malformation. All technical work is done in accordance with standardized techniques employing new techniques such as fluorescence staining. All measurements and analysis are done at Denver with the aid of an automatic chromosome analyzer. A second phase of the study will be concerned with the chromosomal-clinical correlations. The study is in progress.

Honors and Awards: None

Publications: None

Serial No. NDS (CF)-71 PR/PSC 1909
1. Perinatal Research Branch
2. Professional Staff Consultants Section
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Continuation of study of twins born in the Collaborative Study beyond the age of seven years

Previous Serial Number: Same

Principal Investigator: Dr. N.C. Myrianthopoulos, PRB, NINDS

Other Investigators: None

Cooperating Units: All Collaborating Institutions

This study has been discontinued.

Serial No. NDS (CF)-71 PR/PSC 1910
1. Perinatal Research Branch
2. Professional Staff Consultants Section
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Genetics of Obstetric Variables and the Role of Maternal Factors in the Determination of Intelligence and Neurological Performance

Previous Serial Number: Same

Principal Investigators: Dr. A.F. Naylor, PRB, NINDS
Dr. N.C. Myrianthopoulos, PRB, NINDS

Other Investigators: Dr. D. Warburton, College of Physicians and Surgeons of Columbia University

Cooperating Units: Department of Human Genetics and Development, Columbia University

Man Years:

Total:	.50
Professional:	.50
Other:	.00

Project Description:

Objectives: To analyze the variation of obstetric and gynecological factors into heritable and non-heritable components.

Major findings: The completed Record Linkage files have been used to estimate genetic components in obstetric variables, both continuous and discrete: age at menarche, lengths of stages of labor, gestation time, diagonal conjugate, anemia, toxemia, albuminuria, hemorrhage, abnormal presentation, abruptio placentae, fetal death and nine others. A manuscript detailing the results has been drafted and is being revised.

Honors and Awards: None

Publications: None

Serial No. NDS (CF)-71 PR/PSC 1913
1. Perinatal Research Branch
2. Professional Staff Consultant Section
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: The Clinical Significance of Generalized Petechiae at Birth

Previous Serial Number: Same

Principal Investigators: Luz A. Froehlich, M.D., NIAID
Jean G. Oliver, M.A., PRB, NINDS

Other Investigators: Jean S. Nemore, R.N., B.A., PRB, NINDS

Cooperating Units: All Collaborating Institutions

Man Years:

Total:	0.0
Professional:	0.0
Others:	0.0

Current Status:

Evaluation of selected variables did not show any significant differences between cases with significant petechiae and matched controls with respect to speech, language and hearing findings. Other variables should be considered before another evaluation is made. However, eight-year data were not yet available for a significant number of cases at the time of the first evaluation. Therefore, this project has been temporarily suspended until 8-year data collection is completed and in house.

Honors and Awards: None

Publications: None

Serial No. NDS (CF)-72 PR/PSC 1968
1. Perinatal Research Branch
2. Professional Staff Consultants Section
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Prenatal and Postnatal Factors Associated with IQ at Age Four

Previous Project Title: Prenatal and Postnatal Factors Associated with Intellectual Performance at Four Years

Previous Serial Number: Same

Principal Investigators: Sarah H. Broman, Ph.D., PRB-NINDS
Paul L. Nichols, Ph.D., PRB-NINDS
Wallace A. Kennedy, Ph.D., Florida State University

Cooperating Unit: Florida State University

Man Years:

Total:	2.0
Professional:	1.7
Other:	.3

Project Description:

Objectives: The relationship between intelligence at four years of age as measured by the Stanford-Binet Intelligence Scale and a variety of factors related to pregnancy, delivery, postnatal status and social environment was examined among 26,760 children enrolled in the Collaborative Study.

Method: The plan of analysis of these data was exploratory, beginning with an identification of antecedents related to IQ. Following a summary and interpretation of findings, section two presents the history, purpose and methods of the Collaborative Study as a whole. Section three describes the characteristics of the Collaborative Study population and of the sample used in the present analysis. The results of a variable screen in which 169 prenatal, postnatal and environmental factors were correlated with IQ are presented in section four. The 83 variables found to be significantly related to IQ are defined and their distributions in the sample are given. In sections five through 10 the mean IQ for different levels of each predictor variable is displayed within race, sex and social class controls. Significant main effects and interactions are discussed.

The combined effects of the antecedent variables are examined in section 11 in a multiple regression model, and in section 12 in a discriminant function model with subjects divided into low and normal IQ groups. Section 13 presents plans for analyses of the follow-up data on these children at seven years of age. Methodological issues discussed in appended sections include the selection of an outcome variable at four years, the problem of non-linear relationships between antecedent variables and IQ, and the effect of combining data from different Study centers. Final preparation of this report is nearing completion.

Honors and Awards: None

Publications:

Broman, S. H., Nichols, P. L., and Kennedy, W. A.: Precursors of Low IQ in Young Children. In Proceedings of the 80th Annual Convention of the American Psychological Association, Honolulu, Hawaii, September 1-8, 1972, pp. 77-78.

Serial No. NDS (CF)-72 PR/PSC 1969
1. Perinatal Research Branch
2. Professional Staff Consultants Section
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: A study of genetic markers in children of the Collaborative Study

Previous Serial Number: Same

Principal Investigators: Dr. N.C. Myrianthopoulos, PRB, NINDS
Dr. Henry Gershowitz, University of Michigan

Other Investigators: None

Cooperating Units: Collaborating Institutions at Boston, Buffalo, Memphis and Oregon

Project Description:

Objectives: To obtain a distribution of genetic markers in approximately 5,000 children from the Collaborative Study for further genetic analysis.

Methods employed: Blood specimens are being collected in conjunction with the chromosomal study (see NDS (CF)-71 PR/PSC 1908). These are shipped to the Department of Human Genetics of the University of Michigan where the red cells are being separated from the serum and both are frozen in liquid nitrogen.

Proposed course: The frozen samples will be retrieved at a later date and will by typed for a large number of red cell antigens and serum proteins and enzymes. These will be used for the development of an ethnicity index and in a variety of genetic studies.

Current status: To date only about 600 blood specimens have been collected and the prospects of increasing the sample are poor. Plans for utilizing the present sample are being considered.

Honors and Awards: None

Publications: None

Serial No. NDS (CF)-72 PR/PSC 1970
1. Perinatal Research Branch
2. Professional Staff Consultant Section
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Factors Related to Communicative Competence Assessed at the Age of Three Years

During FY 1973 this project was incorporated with Serial No. NDS (CF)-72 PR/PSC 1971.

Serial No. NDS (CF)-72 PR/PSC 1971
1. Perinatal Research Branch
2. Professional Staff Consultant Section
3. Bethesda, Maryland

PHS-NIH
July 1, 1972 through June 30, 1973

Project Title: Biosocial Factors Associated with Communicative Disorders and Competence in Children

Previous Serial Number: Same and incorporating Serial No. NDS (CF)-72 PR/PSC 1970

Principal Investigator: Paul J. LaBenz, Sc.D., PRB, NINDS

Other Investigators: Leo G. Doerfler, Ph.D., Pittsburgh Eye & Ear Hospital
Harry M. Beirne, M.D., Children's Hospital, Buffalo
Luke Gillespie, M.D., Children's Medical Center, Boston
John V. Irwin, Ph.D., Memphis State University
Frank M. Lassman, Ph.D. University of Minnesota
Mrs. Jean G. Oliver, PRB, NINDS
Mr. David Rubinstein, OB, NINDS
Earl D. Schubert, Ph.D., Stanford University
Warren Torgerson, Ph.D., Johns Hopkins University
Peter Workman, Ph.D., University of Massachusetts

Cooperating Units: All institutions participating in the Collaborative Project

Man Years:

Total:	1.3
Professional:	1.2
Other:	.1

Project Description:

Speech, language and hearing (SLH) data have been collected on about 15,000 children at age 3-years, and additional data are being collected at age 8-years. Analysis is being planned to achieve four main objectives:

1. Estimation of the incidence of SLH deficits relative to demographic characteristics of the study population;

2. Selection of the most meaningful and reliable SLH variables and indices to provide an economical descriptor of communicative behavior;

3. Discovery and delineation of relationships between SLH and perinatal variables, especially those relating to neurology;

4. Improvement of SLH methodology for clinical and research applications.

Method:

Data are to be tabulated variously in order to assess the influence of test procedures and demographic factors upon the variance of scores, and to enable estimates of the incidence of communicative disorders as defined operationally. Factor analysis techniques will be applied to derive a minimal set of indices, or a single composite index, which can serve to describe communicative status. Correlation methods will be used to establish relationships with variables of interest. These variables will be drawn from documented areas relating to family history, maternal pregnancy and delivery, neonatal and subsequent examinations, intercurrent health events, and measured physical and mental characteristics.

Current Status:

Preliminary tabulations of 3-year data have been made by items, and by combinations of items, as scored according to protocol criteria and according to revised criteria. Alternative methods of scoring were found not to contribute materially to improved description of communicative behavior. Correlation matrices were constructed to explore relationships among 3-year subtest scores, as well as with several selected variables. Pilot frequency distributions were run on a small sample of 8-year tests prior to large sample tabulations. Plans have been drawn to develop three indices deemed potentially useful. The overall plan of data analysis was revised to focus upon a limited number of key questions.

Publications: None

Honors and Awards: None

Serial No. NDS (CF)-72 PR/PSC 1972
1. Perinatal Research Branch
2. Professional Staff Consultant Section
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Relationships between Spontaneous Speech and Other Test Results at Eight Years of Age

Previous Serial Number: Same

Principal Investigator: Jean G. Oliver, M.A., PRB-NINDS

Other Investigator: Paul J. LaBenz, Sc.D., PRB-NINDS

Cooperating Units: All Collaborating Institutions

Man Years:

Total:	.13
Professional:	.10
Other:	.03

Project Description:

The study of spontaneous speech can provide useful insights into communicative skills, and appears to be a valid index of expressive language proficiency. Samples of spontaneous speech, elicited by visual stimuli (pictures) are being tape-recorded opportunistically at quality control visits where 8-year old children are retested. These samples are later transcribed for analysis. Performances are rated for comparison with data derived from the standard speech, language and hearing battery.

Method:

A tentative set of rating scales has been devised to enable evaluation of several aspects of spontaneous speech output. These include the length of the speech sample as well as the content in terms of quality, organization, elaboration and grammatical structure. Relationships between spontaneous connected speech and a number of traditional measures of communicative ability are to be explored using correlation methods.

Current Status:

Continuation of individual project with eight-year old spontaneous language samples. These samples are being collected and taped at quality control

visits.

Publications: None

Honors and Awards: None

Serial No. NDS (CF)-73 PR/PSC 2053
1. Perinatal Research Branch
2. Professional Staff Consultants Section
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: The Natural History of Congenital Toxoplasmosis: Case Studies from the Collaborative Perinatal Research Project

Previous Serial Number: None

Principal Investigator: Lenore Bajda, M.D., PRB, NINDS

Other Investigators: Dr. John L. Sever, ID, NINDS
M.R. Gilkeson, ID, NINDS

Cooperating Units: Infectious Diseases Branch, NINDS

Man Years:

Total:	.50
Professional:	.50
Other:	.00

Project Description:

Objectives: 1) A search of CRP data for clues to be used as indicators for degree of fetal risk to maternal Toxoplasma infection, 2) To contribute to the prevention of Congenital Toxoplasmosis thru an elucidation of its natural history in the CRP population.

Methods employed: A subsample of 47 gravidae with markedly elevated titers or four-fold rises in Toxo antibody and their 94 control gravidae was provided from an initial CRP population of 23,000 gravidae in the study from onset thru March, 1963 (Sever, J.L. Toxoplasmosis: Serological and Clinical Studies -- Serial No. NDS (CF)-65 PR/ID 1270). This subgroup was subjected to an intensive review of all pertinent original protocols in CRP central files. Maternal characteristics particularly examined in addition to age, race and socio-economic background were prior pregnancy failures, past medical, and current health histories (including drug therapy). Pediatric data reviewed included all medical information available thru the 7 year Neurology examination as well as global IQ scores of the 4 and 7 year Psychological examinations.

Major Findings: Maternal findings included a) a constant exposure to Toxoplasmosis thoughout the child-bearing years, b) higher exposures among Negroes and women born in Puerto Rico, and c) no correlation with urban vs. rural life. Additional findings requiring further support included prior pregnancy failures within 1 year to LMP being greater than in the controls (8.5 to 3.2%) and the

observation that a frequent use of sulfa drugs in the treatment of urinary tract infection during the pregnancy (17%) may have a significant effect on the spectrum of pregnancy outcome. Pediatric findings at a seven year endpoint added to the previously published one-year findings the additional observations of a high rate of "undefined" abnormalities in the case group compared to controls and a high rate of low IQs (24 compared to 17%). The latter finding may become less significant on adjustment for birthweight and degree of maternal education. A larger sample is needed for this analysis.

The implication of the as yet unknown and difficult to measure effect of drug treatment for coexistant medical problems in the Toxoplasma-infected mother demands further study. The spectrum of congenital infection has been expanded to include the possibility of "learning dysfunction" or "disability" in an otherwise "normal" child. This challenge needs urgent attention particularly since Congenital Toxoplasmosis is a preventable disease.

The results of this project were presented at a Symposium on Toxoplasmosis during the XIIIth Congress of the Medical Women's International Association, Paris, France, September 3-8, 1972. Publication is pending.

Honors and Awards: None

Publications: None

Serial No. NDS (CF)-73 PR/PSC 2054
1. Perinatal Research Branch
2. Professional Staff Consultants Section
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Congenital malformations in the Collaborative Study. II. Epidemiology of congenital malformations in multiple births.

Principal Investigators: Dr. N.C. Myrianthopoulos, PRB, NINDS
Dr. C.S. Chung, University of Hawaii

Other Investigators: None

Cooperating Units: None

Man Years:

Total:	.20
Professional:	.10
Other:	.10

Project Description:

Objectives: To study the epidemiology of all congenital malformations in twins and other multiple births in the Collaborative Study, to determine the incidences of these malformations in monozygotic and dizygotic twins and to compare these incidences with those in singletons.

Proposed course and methodology: Extensive use will be made of twin methodology in estimating concordance rates and heritabilities with regard to selected malformations.

Current status: The study is now in progress.

Major findings: None

Honors and Awards: None

Publications: None

Serial No. NDS (CF)-73 PR/PSC 2055
1. Perinatal Research Branch
2. Professional Staff Consultants Section
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Congenital malformations in the Collaborative Study. III. Study of the effects of medical, genetic and socioeconomic factors in the occurrence of congenital malformations

Principal Investigators: Dr. C.S. Chung, University of Hawaii
Dr. N.C. Myrianthopoulos, PRB, NINDS

Other Investigators: None

Cooperating Units: None

Man Years:

Total:	.30
Professional:	.20
Other:	.10

Project Description:

Objectives: To test the etiologic significance of a large number of medical, genetic and socioeconomic factors in the occurrence of major, minor, and selected specific malformations.

Proposed course and methodology: Stepwise regression analysis for 60 selected medical, genetic and socioeconomic variables on all malformations, major and minor malformations, malformations by system and specific malformations with frequency of more than 5/10,000 by race, and using race as a variable. Variables will include multiple birth, maternal age, outcome of previous pregnancies, high blood pressure, diabetes, infections and others.

Major findings: Among several associations found, the most significant and consistent is that of diabetes in the mother with the occurrence of major malformations in the child.

Current status: All major analyses have been completed and a manuscript is being written.

Honors and Awards: None

Publications: None

Serial No. NDS (CF)-73 PR/PSC 2056
1. Perinatal Research Branch
2. Professional Staff Consultants Section
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Congenital malformations in the Collaborative Study.
IV. Maternal diabetes and congenital malformations

Principal Investigators: Dr. C.S. Chung, University of Hawaii
Dr. N.C. Myrianthopoulos, PRB, NINDS

Other Investigators: None

Cooperating Units: None

Man Years:

Total:	.30
Professional:	.20
Other:	.10

Project Description:

Objectives: To explore more fully previous findings suggesting a relationship between diabetes in the mother and the occurrence of congenital malformations in the child.

Proposed course and methodology: Analysis of malformations in children of diabetic mothers and comparison with those of non-diabetic mothers; analysis of possible effects of insulin and the severity of diabetes per se; and analysis of the effects of paternal diabetes.

Major Findings: The incidence of malformations is doubled in children of diabetic mothers compared to that of normal mothers. Malformations associated with maternal diabetes tend to be major, multiple and generalized through systems. There is no effect of paternal diabetes.

Current status: All major analyses have been completed and a manuscript is being written.

Honors and Awards: None

Publications: None

Serial No. NDS (CF)-73 PR/PSC 2057
1. Perinatal Research Branch
2. Professional Staff Consultants Section
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Seizures with Fever, Beginning in the First Year of Life

Previous Serial Number: None

Principal Investigator: Karin B. Nelson, M.D., PRB, NINDS

Other Investigators: Mr. David Rubinstein, OB, NINDS
Miss Eudora Beadle, OB, NINDS

Cooperating Units: All Collaborating Institutions

Man Years:

Total:	1.1
Professional:	1.0
Others:	0.1

Project Description:

All children identified on the one-year diagnostic summary (among 47,222 such summaries) who had had a definite convulsion were evaluated by hand review of charts. Diagnostic reclassification proved necessary. All children whose convulsions were accompanied by fever at onset were evaluated. Perinatal data, subsequent seizure history, 4 and 7 year IQs were examined and several subgroups within the heterogeneous clinical category "Febrile Seizures" identified. The risks of later sequelae was analyzed by subgroup: several clinical features could be isolated which identified children at significantly higher risk for later afebrile convulsive disorders (epilepsy) and mental impairment.

Presentation:

Paper presented at Child Neurology Society Meeting, Ann Arbor, Michigan, October 6, 1972; and by invitation to Western Epilepsy Institute, Salt Lake City, Utah, March 30, 1973.

Honors and Awards: None

Publications: None

Serial No. NDS (CF)-73 PR/PSC 2058
1. Perinatal Research Branch
2. Professional Staff Consultants Section
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Data Analysis Group on Convulsive Disorders

Previous Serial Number: None

Principal Investigator: Karin B. Nelson, M.D., PRB, NINDS

Other Investigators: As yet undetermined

Cooperating Units: All Collaborating Institutions

Man Years:

Total:	0.0
Professional:	0.0
Others:	0.0

Project Description:

An initial meeting to discuss classification of seizure disorders for study purposes, and to plan for utilization of EEG data, is planned. Hand review and classification of case material will be required. Development of plans, procedures, and methods will follow. This effort will be a major area of involvement of the principal investigator, but has only begun at this time.

Honors and Awards: None

Publications: None

Serial No. NDS (CF)-73 PR/PSC 2059
1. Perinatal Research Branch
2. Professional Staff Consultant Section
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Data Analysis Group on Cerebral Palsy

Previous Serial Number: None

Principal Investigator: Karin B. Nelson, M.D., PRB, NINDS

Other Investigators: Robert Groover, M.D., Mayo Foundation, Rochester, Minn.
Alan Leviton, M.D., Children's Medical Center, Boston, Mass.
William Clark, Jr., M.D., University of Oregon Medical School, Portland, Ore.
Jonas Ellenberg, Ph.D., OB, NINDS

Cooperating Units: All Collaborating Institutions

Man Years:

Total:	0.0
Professional:	0.0
Others:	0.0

Project Description:

Participants were selected for the data analysis group on Cerebral Palsy. Definitions were accepted and a format agreed-upon for verification of diagnoses in the relevant case material, with review of definite and possible cerebral palsy cases, as well as those positive for physical findings but bearing no diagnostic coding. Review of 'other' classifications relevant to cerebral palsy diagnoses was planned.

The task of organization for analysis of data on cerebral palsy has only begun as of this writing. It will be the major area of the professional involvement of the principal investigator.

Honors and Awards: None

Publications: None

Serial No. NDS (CF)-73 PR/PSC 2060
1. Perinatal Research Branch
2. Professional Staff Consultants Section
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Birthweight-Gestational Age

Previous Serial Number: None

Principal Investigator: Janet Hardy, M.D.

Other Investigators: Dr. Joseph S. Drage, PRB, NINDS
Dr. Kinne D. McCabe, PRB, NINDS
Dr. Bill H. Williams, PRB, NINDS
Miss Esther Jackson, OB, NINDS
Mr. William Weiss, OB, NINDS
Dr. Mellits, Johns Hopkins University

Cooperating Units: None

Man Years:

Total:	.30
Professional:	.15
Other:	.15

Project Description: During the past year plans have been outlined for achieving the following objectives: 1) Analysis of Collaborative Perinatal Study data relating to birthweight and gestational age, 2) production of a monograph with information pertaining to the above useful for the practicing physician. Plans include the production of frequency and distribution tables of birthweights and gestational ages, and additionally placental weight, body length and other physical characteristics which may be useful in identification of certain categories of infants. Studies will also be done which relate antecedent events (such as maternal disease states, demographic and physical characteristics) to birthweight categories. A Birthweight Index will be developed which will be of some predictive value in relating antecedent events to various birth outcomes. Also, part of the analysis will deal with relating birthweight gestational age categories to various immediate and long term outcomes in children (including mental retardation and cerebral palsy). Hopefully, empirical risk tables can be developed for predictive purposes.

Honors and Awards: None

Publications: None

Serial No. NDS (CF)-73 PR/PSC 2061
1. Perinatal Research Branch
2. Professional Staff Consultants Section
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Physical Growth and Development Task Force

Previous Serial Number: None

Principal Investigator: Kinne D. McCabe, M.D., PRB, NINDS

Other Investigators: Dr. Glen Bartlett, Cleveland Metropolitan General Hospital
Dr. Brigitte de la Burdé, Medical College of Virginia
Dr. Joseph Drage, PRB, NINDS
Dr. Frank Falkner, Fels Institute for Human Development
Dr. Stanley Garn, University of Michigan
Dr. Janet Hardy, Johns Hopkins Hospital
Dr. Richard Osborne, University of Wisconsin
Dr. Robert Reed, Harvard School of Public Health
Dr. T.F. McNair Scott, Children's Hospital of Philadelphia
Dr. Bill Williams, PRB, NINDS
Mr. David Rubinstein, OB, NINDS

Cooperating Units: Children's Hospital University of Buffalo, Medical College of Virginia, University of Oregon Medical School, Children's Hospital of Philadelphia, Brown University

Man Years:

Total:	.45
Professional:	.15
Other:	.30

Project Description: Several projects are included in the work of this Task Force, which is coordinated by the principal investigator. Dr. Bartlett's study involves production of normative distributions, ratios, and correlation coefficients of certain physical growth characteristics of children from birth to seven years. Other task force members are assisting in the production of this data, which will then be used for studying the growth of abnormal children.

Dr. Scott has investigated birthweight gestational age relationships, particularly as related to neonatal mortality. An analysis of the apparent bimodal weight distribution of prematures is planned.

Dr. Garn's work concerns bivariate analysis of growth measurements. These relationships will be studied particularly in relation to socioeconomic status.

Dr. Osborne's study deals with odontometric measurements as a measure of physical growth.

Dr. Falkner's analysis covers growth of twins.

Honors and Awards: None

Publications: None

Serial No. NDS (CF)-73 PR/PSC 2062
1. Perinatal Research Branch
2. Professional Staff Consultant Section
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: The Identification of Types of Minimal Brain Dysfunction and their Relationship to Perinatal, Developmental and Genetic Factors

Previous Serial Number: None

Principal Investigator: Paul L. Nichols, Ph.D., PRB, NINDS

Other Investigators: Ta-chaun Chen, Ph.D., OB, NINDS
Bernard H. Fox, Ph.D., NCI

Cooperating Units: NCI

Man Years:

Total:	.35
Professional:	.25
Other:	.10

Project Description:

Objectives: To discover and describe various types of minimal brain dysfunction and examine their relationship to perinatal, developmental and genetic factors. Methodology for defining sets of outcome variables is being developed.

Honors and Awards: None

Publications: None

Serial No. NDS (CF)-66 PR/P 1345
1. Perinatal Research Branch
2. Section on Pathology
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Influencing Factors in Sudden Unexpected Death

Previous Serial Number: Same

Principal Investigators: Toshio Fujikura, M.D.

Other Investigators: Joseph S. Drage, M.D.

Cooperating Units: All Collaborating Institutions

Man Years:

Total:	0.15
Professional:	0.10
Others:	0.05

Project Description:

The study has been revised with new additional cases of sudden infant death syndrome (SIDS). The purpose of this paper is to report the analysis of our SIDS cases and to evaluate the epidemiologic factors, particularly the incidences according to the number of single births and autopsied deaths.

The incidence of SIDS over the age of seven days and up to seven years was 0.34% or 137 of 39,773 single births in a prospective study. Since these SIDS cases included autopsied deaths only, the incidence seemed to be conservative. Although the incidences for Negro and male births were higher than those for the white and female respectively, such Negro and male preponderance disappeared if the incidence was based on autopsied deaths. About half of infant deaths within one to four months of age were classified as SIDS and the peak incidences were within two and three months (55.5% and 57.8%). This suggests that infants in the age group may have a general character which is susceptible to sudden and unexpected fatal outcome in addition to other conditions. The findings of epidemiologic factors such as birthweight, gestational age, socio-economic index and month of death were not impressive or negative among autopsied deaths in contrast with reported data.

The paper is being prepared for publication.

Honors and Awards: None

Publications: None

Serial No. NDS (CF)-69 PR/P 1772
1. Perinatal Research Branch
2. Section on Pathology
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Experimental Ischemia of the Spinal Cord: Histological Studies After Anterior Spinal Artery Occlusion

Previous Project Title: Pathologic Effects of Ligation of the Anterior Spinal Artery and/or the Great Radicular Artery in Monkeys

Previous Serial Number: Same

Principal Investigators: Oscar Aparicio, M. D.

The investigator for this study has left the Perinatal Research Branch and we have not been able to contact him. Study is discontinued.

Serial No. NDS (CF)-70 PR/P 1859
1. Perinatal Research Branch
2. Section on Pathology
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Mental and Motor Development in Monozygotic Co-Twins with Dissimilar Birthweights

Previous Serial Number: Same

Principal Investigators: Toshio Fujikura, M.D.
Luz A. Froehlich, M.D.

Other Investigators: None

Cooperating Units: All Collaborating Institutions

Man Years:

Total:	0.0
Professional:	0.0
Others:	0.0

Project Description:

Developmental measures in 125 monozygotic twin sets with unequal birthweights between co-twins were studied. There were no significant differences between co-twins in the Bayley mental and motor scores at eight months nor the Stanford-Binet I.Q. at four years. A reportedly higher I.Q. for the heavier monozygotic twins was not confirmed in this study, even among pairs with large birthweight differences (mean differences 26-28%). Although the effects of nutrition on the mental development of the fetus are currently of great concern, these data suggest that the developing human brain seems to have a strong resistance to intrauterine deprivation.

The paper has not been approved for publication. Because interpretation of the data is controversial, the future of this manuscript is still pending.

Honors and Awards: None

Publications: None

Serial No. NDS (CF)-71 PR/P 1911
1. Perinatal Research Branch
2. Section on Pathology
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: A Follow-Up of Children with Single Umbilical Artery

Previous Serial Number: Same

Principal Investigators: Toshio Fujikura, M.D.
Luz A. Froehlich, M.D.

Other Investigators: None

Cooperating Units: All Collaborating Institutions

Man Years:

Total:	0.0
Professional:	0.0
Others:	0.0

Project Description:

Single umbilical artery (SUA) is known to be frequently associated with other congenital malformations and to be as one of the most common malformations in the human. Other significant findings are a 25 percent rate of low birthweight, a consistently higher frequency of SUA among whites than among Negroes, and a high association with diabetes and velamentous insertion of umbilical cord. These previous studies have all been on newborns and perinatal deaths and no follow-up studies have appeared in the literature.

In whites and Negroes of our Collaborative Research Study, there were 344 infants born with SUA. Of the 344 single births, 48 (14 percent) were stillbirths, neonatal and infant deaths. The remaining 296 are presumed to be living and 266 had careful follow-ups up to four years of age. The incidence of single umbilical artery (SUA) was 0.9 percent in general and was higher in whites (1.2 percent) than in Negroes (0.5 percent). The incidence of SUA survivors was still 0.7 percent in spite of high mortality rate (14 percent).

Associated malformations were 19 of 36 SUA deaths (52.8 percent). Cardiovascular anomalies were less involved in the SUA group, compared to the total group of malformed deaths. Anomalies involving other systems were comparable in the two groups.

In the follow-up study comparing 266 SUA survivors with 798 matched controls, mean values of body weight, body length, and head circumference at birth, four months, one year and four years were recorded, as well as motor and mental

scores at eight months and I.Q.'s at four years. These values between the two groups were almost equal at each developmental stage. The results of the one-year neurological, four-month pediatric, and eight-month psychological examinations were reviewed in the two groups. The incidence of definite abnormal conditions was about equal in the two groups, except for several malformations such as inguinal hernia which were higher in the SUA group. This indicates that in the absence of associated severe malformations, which were common among the stillbirths and neonatal deaths, SUA survivors developed just as normally as infants without SUA.

The paper was presented at the joint meeting of American Association of Pathologists and Bacteriologists (69th Annual Meeting) which was held in Cincinnati, Ohio from March 11 to 14, 1972.

The paper is being prepared for publication.

Honors and Awards: None

Publications: Fujikura, T. and Froehlich, L.A.: A Follow-Up of Children with Single Umbilical Artery. Pediatrics. In press.

Serial No. NDS (CF)-71 PR/P 1912
1. Perinatal Research Branch
2. Section on Pathology
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Birthweight, Gestational Age and Renal Glomerular Development as Indices of Fetal Maturity

Previous Project Title: Birthweight in Relation to Renal Glomerular Development and Gestational Age in Whites and Negroes

Previous Serial Number: Same

Principal Investigators: Toshio Fujikura, M.D.
Luz A. Froehlich, M.D.

Other Investigators: None

Cooperating Units: All Collaborating Institutions

Man Years:

Total:	0.0
Professional:	0.0
Others:	0.0

Project Description:

Birthweight, body length and X-ray evaluation of bone development continue to be widely used as measures of fetal maturity. However, these are factors of somatic growth and are not necessarily directly related to growth and maturity of internal viscera. On the conviction that histologic examination of an organ would give a more accurate picture of the maturation of internal viscera, a histologic evaluation of the kidneys of 514 neonatal deaths and fresh stillbirths was undertaken. Below and up to 34 weeks gestation in females and 35 weeks gestation in males, the mean birthweights in Negroes were consistently higher than in whites. Above these gestational ages the mean birthweights of whites exceeded those of Negroes. In spite of such birthweight differences, renal maturity measured by examining the nephrogenic zone of early neonatal deaths and stillbirths was not significantly different in the two races within each gestational age interval. The concept that Negro fetuses mature more rapidly appears invalid. Gestational age seems to be better as an indicator of fetal maturity than birthweight if gestational age is properly recorded. Study has been completed.

Honors and Awards: None

Publications: Fujikura, T. and Froehlich, L.A.: Birthweight in Relation to Renal Glomerular Development and Gestational Age in Whites and Negroes. Am. J. Obstet. Gynecol., 113:627-631, 1972.

Serial No. NDS (CF)-72 PR/P 1973
1. Perinatal Research Branch
2. Section on Pathology
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Renal Glomerular Development in Infants of Diabetic Mothers

Previous Serial Number: Same

Principal Investigators: Toshio Fujikura, M.D.

Other Investigators: Shirley Driscoll, M.D.

Cooperating Units: Boston Hospital for Women
Lying-In Division
221 Longwood Avenue
Boston, Massachusetts 02115

Man Years:

Total:	0.15
Professional:	0.10
Others:	0.05

Project Description:

Our preliminary result showed that in maternal diabetes 89.7% of infants between 35-37 weeks gestation still present evidence of a nephrogenic zone where only 62.5% is expected. Since most of these infants were delivered by elective Cesarean section, it would perhaps have been beneficial to the baby had delivery been postponed for another one or two weeks to allow more complete maturation of kidneys and maybe other organs as well.

Delayed visceral maturation could be an underlying course of the morbidity in infants of diabetic mothers. In order to confirm statistically this interesting result, 130 cases of neonatal deaths (up to three days old) and fresh still-births whose mothers are known to be diabetic in the Joslin Clinic have been used for the evaluation of renal glomerular development.

Honors and Awards: None

Publications: None

Serial No. NDS (CF)-72 PR/P 1974
1. Perinatal Research Branch
2. Section on Pathology
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: The Significance of Meconium Staining in Neonatal Deaths

Previous Serial Number: Same

Principal Investigators: Toshio Fujikura, M.D.

Other Investigators: None

Cooperating Units: All Collaborating Institutions

Man Years:

Total:	0.25
Professional:	0.10
Others:	0.15

Project Description:

Meconium staining of the amniotic fluid is one of the classic signs of fetal distress in utero. However, in 12 to 25% of deliveries complicated by fetal meconium passage, no definite cause of such conditions is found. The significance of meconium stained fluid is still a widely debated subject. The purpose of this study is to investigate pathologic findings of neonatal deaths with the passage of meconium and to confirm the significance of this condition relating to hyaline membranes, atelectasis, cardiovascular malformations and erythroblastosis.

The incidence of meconium staining of the placental membranes, umbilical cord and/or fetal body was 10.3% in 42,490 live births and 18.1% in 788 neonatal deaths. The presence of meconium staining was significantly high in birthweight intervals above 3,501 grams. The incidence of hyaline membranes and atelectasis was naturally lower in the stained group (143 cases) than in the nonstained group (645 cases). However, this trend was still present in premature neonatal deaths below 2,500 grams. Aspirated meconium (bile pigments) is considered as a strong surfactant and such surface active substance may prevent production of atelectasis and hyaline membranes in the neonatal period. Erythroblastosis was four times higher in the stained group and the presence of meconium staining may be considered as the dangerous sign of kernicterus in relation to hypoxia and high blood bilirubin. Cardiovascular malformations were twice higher in the stained group and should be checked carefully at birth.

Pneumonia and pulmonary hemorrhages were not significantly higher in the stained group, but severe placentitis was present predominantly in this group.

Maternal factors such as abruptio placentae, placenta previa, prolapsed cord, prolonged ruptured membranes and diabetes were not different between the two groups. Pre-eclampsia with or without hypertension was twice higher in the stained group as previously reported.

The presence of meconium staining does not imply that the fetus is in extreme distress, rather that it can be considered as a marker for hypoxia.

The paper is being prepared for publication.

Honors and Awards: None

Publications: None

Serial No. NDS (CF)-72 PR/P 1975
1. Perinatal Research Branch
2. Section on Pathology
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Organ Weight Change of Infants with Pulmonary Hyaline Membranes

Previous Serial Number: Same

Principal Investigators: Toshio Tujikura, M.D.

Other Investigators: None

Cooperating Units: All Collaborating Institutions

Man Years:

Total:	0.15
Professional:	0.05
Others:	0.10

Project Description:

Organ weight/brain weight ratios (O/B ratios) remain practically constant for each developmental stage in infants weighing 1,000 to 3,500 grams and organ weights seem to increase proportionately at constant rates during the perinatal period. Subtle weight changes in internal organs are detected by using O/B ratios. The aims of this study are to investigate O/B ratios in relation to pulmonary hyaline membranes (PHM) or pneumonia and to discuss the possible etiology of hyaline membrane disease.

In 227 neonatal deaths less than three days old, O/B ratios were separately calculated in the three groups: pulmonary hyaline membranes (84 cases), pneumonia (67 cases) and controls (76 cases). Their diagnoses were mutually exclusive. In the group of PHM the lungs were excessively heavy, indicating the presence of pulmonary edema, congestion, hemorrhage and possible heart failure. The thymus was also large in the PHM group. However, weights of other organs: adrenal glands, spleen, heart, kidneys and liver, in both PHM and pneumonia groups were not significantly different from the counterparts of controls.

Since these pulmonary findings, which have been reported with hyaline membrane disease, are difficult to assess quantitatively by microscopic examination, the heavy weight may indicate the serious condition of fluid retention in the lung.

The paper is being prepared for publication.

Honors and Awards: None

Publications: None

Serial No. NDS (CF)-72 PR/P 1976
1. Perinatal Research Branch
2. Section on Pathology
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: The Effects of Profound Hypothermia in Newborn Mice: Prolonged circulatory arrest without cerebral damage

Previous Serial Number: Same

Principal Investigators: Toshio Fujikura, M.D.

Other Investigators: None

Cooperating Units: None

Man Years:

Total:	0.4
Professional:	0.2
Others:	0.2

Project Description:

Objectives: The purpose of this experimental study is to test the tolerance limits of profound hypothermia in newborn mice and to determine the somatic and motor development, especially neurological abnormalites of surviving mice.

Method Employed: Newborn mice at birth are still premature and are unable to properly control their body temperature. In each delivery the litter was divided equally into two groups. Half of the litter served as controls. Within 24 hours of age the other half of the litter was placed in a refrigerator at 7°C temperature for 6 hours, 12 hours and 18 hours respectively. After cooling, exposure treated animals were kept at room temperature (24°-25°C) for 5-6 hours and the survivors were returned to their mothers. In order to permit maximal evolution of any central nervous system injury, survival for two months was attempted in all animals. All animals were killed at two months of age. The brain and spinal cord were examined grossly and microscopically.

Major Findings: Total circulatory and respiratory arrest followed within 1 hour of cooling. All of the animals after 6 hours and 12 hours respectively of hypothermic anoxia revived spontaneously within 20 minutes of rewarming. As compared to the control litter, the two-month old treated survivors appeared normal without neurological deficits. Body weight and body length were within normal range at that age. This suggests that newborn mice and their brains can tolerate a longer period of anoxia than was formerly expected under profound hypothermia.

Significance to Biomedical Research and the Program of the Institute:
The clinical application of profound hypothermia in heart surgery on newborn infants is winning wide support in the United States and other countries. The findings of the present study encourage the feasibility of profound hypothermia for a limited period, during which time no brain damage is expected in the infants even after circulatory arrest.

The paper is being prepared for publication in the Biology of Neonate.

Honors and Awards: None

Publications: None

Serial No. NDS (CF)-73 PR/P 2063
1. Perinatal Research Branch
2. Section on Pathology
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Genesis of Single Umbilical Artery

Previous Serial Number: None

Principal Investigators: Kenichiro Ezaki, M.D.*

Other Investigators: Toshio Fujikura, M.D.
Takashi Tanimura, M.D.*

Cooperating Units: Department of Anatomy, Faculty of Medicine,
Kyoto University, Kyoto, Japan
*The faculty member

Man Years:

Total:	0.0
Professional:	0.0
Others:	0.0

Project Description:

It is generally accepted that Single Umbilical Artery (SUA) is the result of either aplasia or secondary atrophy of the missing umbilical artery. In order to investigate this mechanism, 1544 umbilical cords of human embryos (Streeter's Horizons 14-23) were examined carefully.

The frequencies of SUA were: one in 1471 externally normal live embryos, one in 31 externally dead embryos, two in 31 malformed live embryos, and one in 12 malformed dead embryos. The incidence of SUA in these embryos was lower than that in newborns. This finding seems to be against the theory of aplasia.

In several cases, one umbilical artery was considerably smaller than the other in the caliber. This indicates that SUA could be produced by secondary atrophy of one of the umbilical arteries.

This paper was presented at the Symposium on Congenital Anomalies and Malignant Tumors, held in Nügata, Japan, July 13-14, 1972, and an abstract appears in Teratology, 6:105, 1972. Study has been completed.

Honors and Awards: None

Publications: None

Serial No. NDS (CF)-73 PR/P 2064
1. Perinatal Research Branch
2. Section on Pathology
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Blood Flow Study in the Intervillous Space:
Sickling of maternal erythrocytes in the placenta

Previous Serial Number: None

Principal Investigators: Toshio Fujikura, M.D.

Other Investigators: None

Cooperating Units: All Collaborating Institutions

Man Years:

Total:	0.50
Professional:	0.30
Others:	0.20

Project Description:

Objectives: The purpose of this study is to investigate the blood circulation in the intervillous space using maternal erythrocytes with or without sickling as a tracer.

Methods Employed: About 100 placentas of maternal sicklemia case have been selected for this investigation. In placental sections evaluated, 3 microscopic fields; subchorionic space, intralobular space and decidual veins are chosen. The number of maternal erythrocytes with or without sickling are counted in each field. Two cross sections derived from placental body and margin are evaluated in each case.

Major Findings: (1) Percentage of sickling in subchorionic space is 10-20% only. This indicates that oxygenated blood enters basally and is expelled upwards towards the chorion. It is not necessary to bath the fetal cotyledons on the way. Even in the subchorionic space of placental margin, percentage of sickling is below 20%. (2) Percentage of sickling in intralobular space is from 80% to 100%. (3) Percentage of sickling in decidual veins is from 30% to 60%. Considerable contamination between oxygenated and non-oxygenated blood is common in the circulation.

Proposed Course: Completion of this study during next fiscal year.

Honors and Awards: None

Publications: None

Serial No. NDS (CF)-73 PR/P 2065
1. Perinatal Research Branch
2. Section on Pathology
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Placental Transmission of Sickling Erythrocytes from Mother to Fetus

Previous Serial Number: None

Principal Investigators: Toshio Fujikura, M.D.

Other Investigators: None

Cooperating Units: All Collaborating Institutions

Man Years:

Total:	0.50
Professional:	0.30
Others:	0.20

Project Description:

Objectives: The purpose of this study is to investigate materno-fetal passage of maternal erythrocytes showing sickling in the placenta. Transplacental passage of cells must be considered in an important immunologic context, that of graft vs. host reactions.

Methods Employed: If the presence of sickling in the fetal circulation of the placenta is noted, the number of sickled erythrocytes are counted in 500 erythrocytes. Sickled erythrocytes are examined either in the umbilical vein or fetal veins of the chorion. About 100 placentas of maternal sicklemia case have been selected for this investigation.

Major Findings: Since sickling of fetal erythrocytes is not commonly found under normal venous oxygen tension, sickled erythrocytes in the fetal circulation is supposed to originate from maternal erythrocytes. Sickled erythrocytes pass the placental barrier from mother to fetus and the incidence of the positive finding in the fetal circulation is 100% so far. The frequency of sickled erythrocytes is about 5-10 or 1-2% in 500 erythrocytes. In several cases syncytial sprouts are also noted in the fetal circulation.

Significance to Biomedical Research and the Program of the Institute: Placental passage of maternal erythrocytes and lymphocytes from mother to fetus is extremely higher than is formerly expected in the placental barrier. Immunologically competent maternal cells cannot only cross the placenta, but also permanently colonize and function in man. If so, the relation of certain disease processes to such materno-fetal chimerism deserves close attention.

Honors and Awards: None

Publications: None

Serial No. NDS (CF)-73 PR/P 2066
1. Perinatal Research Branch
2. Section on Pathology
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: The Influence of Birth Rate on Neonatal Mortality by Birth Weight, Gestational Age, Race and Sex

Previous Serial Number: None

Principal Investigators: Toshio Fujikura, M.D.

Other Investigators: Charles Brown, Ph.D.
Biometry Branch, NCI

Cooperating Units: All Collaborating Institutions

Man Years:

Total:	0.4
Professional:	0.10
Others:	0.30

Project Description:

Objectives: The purpose of this study is twofold. The first is to investigate the relationship of neonatal death rate with birth weight and gestational age. The second is to demonstrate the effect of birth rate in relation to sex and race upon the mortality relative to birth weight and gestational age.

Major Findings: Neonatal mortality rates between Caucasians and Negroes, between males and females were compared with the corresponding birth rates (percentage distributions) in the birth weight and gestational age intervals. The mortality rate in the individual interval was inversely related to the corresponding birth rate. This inverse relationship was significantly found in the premature births ranging from 1601 grams to 2600 grams of birth weight, or from 28 weeks to 37 weeks of gestation. This suggests that the incidence of lethal premature problems such as pulmonary hyaline membrane disease should be calculated by using perinatal or neonatal deaths as well as all or live births.

Significance to Biomedical Research and the Program of the Institute:
Live births born in a given birth weight or gestational age interval may not be as homogeneous as expected statistically with a large number of births. This inverse relationship between perinatal or neonatal mortality and birth rates (percentage distributions of all or live births) is also found in a large population such as U.S. vital and health statistics. It is not necessary to be associated with a small number of the sampled population.

The paper is being prepared for publication in the American Journal of Obstetrics and Gynecology.

Honors and Awards: None

Publications: None

Serial No. NDS - 63 OD/OPE 1144
1. Office of the Director
2. Office of Planning & Evaluation
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: An Instrument For The Conduct of a Retrospective Study of Seizures, Cerebral Palsy, Mental Retardation and Other Neurological and Sensory Disorders of Infancy and Childhood.

Previous Serial Number: NDS (CF)-63 PR/OC 1144

Principal Investigators: Z. A. Shakhashiri, M.D., PRB, NINDS
Leonard V. Phelps, Clearwater, Florida
Glen S. Bartlett, M.D., Case Western Reserve Univ., Cleveland, Ohio

Other Investigators: Lenore Bajda, M.D., PRB, NINDS
John R. Day, M.D., Chevy Chase, Maryland
Blanche L. Vincent, S.N.O., Greensboro, North Carolina
Zula C. Meekham, B.S.N., PRB, NINDS
Rose R. Tortorella, PRB, NINDS

Cooperating Units: Georgetown University Hospital, Retarded Children's Clinic, Selected Maternity Hospitals and Physicians in Metropolitan Washington.

Man Years:

Total:	.35
Professional:	.30
Other:	.05

Project Description:

Objectives: Design an instrument for the conduct of a retrospective study of seizures, cerebral palsy, mental retardation and other neurological and sensory disorders of infancy and childhood in order to test certain basic and important hypotheses concerning the occurrence of neurological damage.

Methods employed: Recognized damaged outcomes of pregnancy, such as seizures, diplegias, hemiplegias and choreoathetoids are to be studied and related to defined perinatal or postnatal events. These outcomes were selected because they were construed to be related to or manifestations of or involved in the biological or psycho-sociological mechanism underlying the following hypotheses: (1) anoxia, (2) toxic influences on the brain, (3) metabolic influences, (4) trauma to the head, (5) infection of the brain, (6) dehydration of the child, (7) genetic or familial patterns, (8) socioeconomic status, (9) prematurity, and (10) nutrition.

This study will be prepared in three manuscripts, one to portray biological and socioeconomic correlates of Mental Retardation, one to portray biological and socioeconomic determinants of Mental Retardation, and one to portray the comparative methodology of two record-anchored studies focusing on (1) routine hospital records, and (2) research-designed hospital records.

Honors and Awards: None

Publications: None

Serial No. NDS - 63 OD/OPE 1146
1. Office of the Director
2. Office of Planning & Evaluation
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Public Health Implications Study of Perinatal Mortality in the Collaborative Study and in the Collaborative Study Cities.

Previous Title: Revision and Expansion of Previous Project Entitled A Commentary On The Appropriateness of the Use of Certain Tabular Data, For Formulating Generalizations Concerning Populations In the Same Cities as Those In Which The Collaborative Study On Cerebral Palsy, Mental Retardation And Other Neurological and Sensory Disorders of Infancy And Childhood Is Being Conducted.

Previous Serial Number: NDS (CF)-63 PR/OC 1146

Principal Investigators: Z. A. Shakhashiri, M.D., PRB, NINDS
Leonard V. Phelps, Clearwater, Florida
Glen S. Bartlett, M.D., Case Western Reserve Univ., Cleveland, Ohio

Other Investigators: None

Cooperating Units: The Census Bureau and the National Center for Health Statistics cooperated in the furnishing of necessary statistical information for the United States and cities. The respective state or city health departments provided natality and mortality data for the Project.

Man Years:

Total:	0.50
Professional:	0.30
Other:	0.20

Project Description:

Objectives: To evaluate fetal and infant mortality of the Collaborative Study population and of the cities from which that population is drawn, with the aim of comparing the two populations, city by city, and institution by institution, on mortality characteristics.

Methods Employed: In addition to the data previously available from the National Center for Health Statistics for perinatal events, detailed data on natality and perinatal mortality were sought for the study cities from

either the state or city health departments, whichever had jurisdiction for these records. The data include figures for livebirths, stillbirths, and deaths under 24 hours, 1 day to 7 days, 8 days to 28 days and 1 month to 12 months, evaluated by birth weight, length of gestation, race and sex, and plurality for the years 1959 through 1966. Corresponding data were compiled by institution for the PRB study population. The state or city health departments furnished either completed tabulations or raw data to be tabulated.

Considerable effort has been expended to create a usable data file of the external data being obtained in connection with this study. The aim is to provide a file with more general utility than the limited scope of this study. When such a file is created, the information necessary to make use of the file will be made available to interested persons.

Major Findings: Evaluation of birth weight and length of gestation data for all core live births (first and subsequent pregnancies) reveals differences between races and between sexes that are generally persistent among all the institutions. Birth weights are lighter among non-whites than among whites, and among females than among males. With white males the heaviest, the order of decline is next white female, then, at about the same weight, non-white males, then non-white females. There is a particular excess of non-whites at low birth weights (2500 grams or less). Length of gestation is shorter among non-white than among whites by about 1 week, with an excess of both short-gestation and long-gestation deliveries among non-whites. Length of gestation is slightly shorter among males than among females in both races, but less consistently so among whites.

Perinatal mortality has declined in the study population since its first year, with a transient elevation in 1962. Group I mortality (fetal deaths 1001 grams and over plus deaths under 7 days of age per 1000 total births) declined from 28.5 in 1959 to 17.6 in 1966 (mean 21.7) among whites and from 33.2 to 21.8 (mean 28.4) among non-whites. Group II mortality (fetal deaths 501 grams and over plus deaths under 28 days per 1000 total births) declined from 34.9 in 1959 to 19.1 in 1966 (mean 26.0) among whites and from 39.8 to 25.5 (mean 33.3) among non-whites. These trends tended to persist from institution to institution, though to varying degrees.

First phase: A manuscript is in preparation, describing birth weight, fetal, neonatal and infant mortalities of the COLR population during 1959-1966 with delineation of trends and suggested interpretations. Second phase: The mortality trends during 1959-1966 of the populations of the cities where the COLR centers are located shall be delineated and interpreted. Third phase: The mortality trends of the two populations in Phases one and two shall be compared and interpreted.

Honors and Awards: None

Publications: None

ANNUAL REPORT

July 1, 1972 through June 30, 1973

Special Programs Branch
Collaborative and Field Research
National Institute of Neurological Diseases and Stroke
National Institutes of Health

The Special Programs Branch of C&FR, NINDS was established in July 1971 as a result of moving certain direct contract operations out of the Office of the Director, NINDS.

The Special Programs Branch has the responsibility for:

1. The development, conduct, and direction of programs for scientific and technical information exchange relevant to the neurologic and sensory sciences on a national and international basis.

2. Coordination of collaborative research and development studies with Institute goals and objectives carried out in foreign laboratories with U.S. government support under Public Law (P.L.) 480.

3. Development and conduct of national and international directed research programs of relevance to the Institute's goals and in response to Institute program needs and priorities as identified by the Director, NINDS and the Associate Director, C&FR, NINDS.

Early in the year the Special Programs Branch moved from Building 36 to the Federal Building, which resulted in more consolidated space. However, there is some disadvantage at being removed from the proximity of the National Library of Medicine (NLM) and the NIH Library as the activities of the Scientific Information activities of the Branch call for considerable work in these facilities.

There was one staff change: Mrs. Lillian Kaplan left the Special Programs Branch, to join the Information Analysis Branch of the National Institute of Child Health and Human Development. This is a considerable loss, as Mrs. Kaplan has been instrumental in interactions with GPO and other outside scientific information services. She has not been replaced and her work has to be distributed among the others. This may cause termination of some of the in-house activities related to the Scientific Information Programs.

The Scientific Information Programs of the Special Programs Branch, particularly the Neurological Information Network (NIN) has continued to have a

growing impact on the NINDS community of users.

The NIN at present consists of the central office in the Special Programs Branch, the Brain Information Service (BIS) at UCLA, the Information Center for Hearing, Speech, and Disorders of Human Communication (ICH&DHC) at Johns Hopkin University Medical School, the Clinical Neurological Information Center (CNIC) at the University of Nebraska, and the Cerebro Vascular Alerting Service (CVD) at the Mayo Foundation. Close interaction with the National Library of Medicine is also maintained as part of the NIN. The detailed activity of these centers is given in the contract narratives.

The impact of the NIN can be seen by the increasing demand for services from all of the specialized centers, special studies to evaluate center products, the increasing amount of interaction between the information centers with professional societies, and the international recognition of the NIN.

The BIS currently provides service to about 6000 scientists on a regular basis (monthly or semi-monthly) and during the year another group about the same size queried them and received irregular services of some kind making a total of about 12000 scientists to whom they provided services. The Center for Hearing, Speech and Communication has also increased their audience, circulating an announcement of service to all of the members of the American Speech and Hearing Association which has produced on alarming number of inquiries. Twelve thousand copies of variou prepared bibliographies were sent out from written requests.

Three evaluation studies of NIN products are being carried out. One has been completed on the Neurosurgical Biblio-Index and the report is in draft form. Two others on the Parkinson's Disease and Related Disorders alerting bulletin, and another on Biogenetic Amines an alerting bulletin,are underway.

The Neurosurgical Biblio-Index is a recurring bibliography designed for neurosurgeons. It is produced quarterly from the National Library of Medicine MEDLAS system and distributed to the 6000 subscribers of the Journal of Neurosurgery. This stu has indicated that a recurring specialized bibliography sent to individuals is indeed a valuable and useful tool, and while people will pay a modest price for it, it would be difficult to recover all the production costs, i.e. the effort of NLM. Preliminary results from the evaluation of the other two products (literature alerting bulletins) indicate that they have a significant effect on the direction of scientists thinking and direction of research by calling attention to articles they had not seen.

The professional societies in the many areas served by the NIN are increasingly coming to the centers for help in problems of scientific communication.

The Society for Neurosciences has recognized the importance of the Brain Information Service as the information service for the neurosciences. They are working with BIS in developing a new format for the annual meeting program and for consultation on other problems. The Director of the Brain Information Service and the Chief of the Special Programs Branch, NINDS are members of the Society for Neuroscience Committee on Scientific Communication so that there is feedback from and interaction with the user community of BIS to develop services which best meet the needs of the neuroscientists.

The Academy of Ophthalmology and Otolaryngology has asked the Information Center for Hearing, Speech, and Disorders of Human Communication to develop a series of special meetings on topics of special interest. These meetings are to be funded by the Society with the ICHS&DHC producing the proceedings.

The impact of the NIN is also evident from the recognition it has received outside the borders of the United States. The British Office of Scientific and Technical Information is exploring the establishment of a satelite service to distribute the products of the Center for Hearing, Speech, and Disorders of Human Communication to the appropriate scientists of the United Kingdom. The Scientific Information Center in Brazil has been in contact with both the Information for Hearing and Speech Center and the Brain Information Service to develop a cooperative effort. There have been some informal discussions for exchange of effort with the ministers for Scientific Information of Poland and in Egypt. The World Health Organization is interested in a possible NIN expansion. The international professional societies in Neurosurgery, Neurology, and Brain Research are all interested in having the NINDS Neurological Network expand worldwide. This kind of expansion, while very exciting, has to be limited because of budgetary constraints, but it, nonetheless, indicates the impact of the neurological information network.

The Scientific Information Program's Advisory Committee (SIPAC) is undertaking an in-depth review of these programs to try to develop priorities for continuation, preservation, or alteration of the program in view of the severe budgetary restrictions we are facing.

The Clinical Information Center at Nebraska has been underway for only about one year and they are already beginning to draw considerable notice from the clinical community. They have a National Advisory Committee drawn from the leaders in neurology and neurosurgery. They have produced an innovative new alerting bulletin

called "Concise Neurological Reviews" in which papers on the same subject are grouped together and the current claim of each paper listed and referenced. The citations are then given as a separate section. Reading this bulletin allows one to get a feel for current work going on in various areas. If more information is required the reference is there. It has been covering some two or three hundred articles a month, an amount of reading almost impossible for any one person to do and do any other work as well. This is a true innovation in the field of dissemination and scientific informatic

The Cerebrovascular Disease Alerting Service abstracts which appear in the journal Stroke have been put on tape for computer search and retrieval. This, along with Epilepsy Abstracts form the nucleus for an eventual large scale on-line information and retrieval system.

The Central Office of the Neurological Information Network in the Office of the Special Programs Branch has continued to produce Parkinson's Diseases and the Related Disorders, a alerting bulletin of relevant citations which comes out monthly. I at present is the subject of an evaluation study.

A directory of sources of information regarding neurosciences is being developed by the Special Programs Branch. This will include the major sources of information about the neurosciences with the exception of monographs and textbooks. This will be in the order of about 80 pages in length. The Society for Neurosciences wants to distribute this to all its members. The English Brain Research Society has aske for about 1000 copies and several other societies have asked for enough copies so that their officers can evaluate for distribution among their memberships. It will probably b available to other scientists through GPO but how it will be sold has not yet been resolved.

The central office of the network also has continuing interacting with othe information systems not only in the government but outside the government.

The Special Programs Branch has maintained a collection of citations of case reports of neurological diseases and review articles of neurological problems whic appear in literature and maintain these in a file which has as its major sections the thirteen large program areas of the institute. This collection is generated by a search the new entries into the MEDLARS once a month. These citations and their index terr are printed on 3X5 cards. These are sent to the Special Programs Branch where they ar reviewed and entered into the file.

With the advent of the Clinical Neurological Information Center a duplica set of cards has been sent to the Clinical Information Center in Nebraska for their use making this information more widely available to the neurological community.

At present the file contains several thousand case reports on over 800 disease entities and syndromes. Most of these case reports are lost because they do not get put into the Index Medicus, although they are in the MEDLARS file. By isolating the case reports in a special file we have a resource which will rapidly respond to questions about unusual diseases or find the latest review of a disease problem if required.

Recently the searches have been done on Medline, and these are proving unsatisfactory. Also the NLM has stopped using card format as printout and unless the problems can be resolved the project will probably be terminated.

Because of personnel and budget cuts the file will probably only be maintained by the Clinical Neurological Information Center.

A special workshop on devices to detect respiratory difficulty was held by the Special Programs Branch at the request of the Director of NINDS. Dr. Joseph Ogura, Professor and Head of the Department of Otolaryngology, at the Washington University in St. Louis, when he was a member of the NINDS council raised the question of the technological possibilities for developing devices to detect respiratory obstruction or difficulty in patients with tracheostomy. A preliminary survey suggested that there were several people working on this problem and some devices existed. The workshop was held on July 20, 1972 in St. Louis, Missouri.

The problem of concern was outlined by Dr. Ogura who stressed that this meeting was concerned, not with measurements of respiration in general, respiratory difficulty in general, or respiratory physiology, but only the specific clinical problem of the earliest possible detection of difficulty developing in a patient with a tracheostomy.

What was desired was a system which would sound an alarm and call attention of the nursing staff or other attending aids, that the patient was beginning to develop trouble, and something needed to be done. The various research groups working on this problem presented their work and/or plans. The Ames Research Center in Moffett Field, California have developed an Apnea detector; a team from Johns Hopkins University presented their plans to attack this problem; a group from Utah presented a plan to attack the problem and the group from Colorado presented their program.

There was a good deal of general discussion which developed the concensus that with the ongoing work there were devices being developed which did meet the problem outlined by Dr. Ogura.

Exchange of information between the various investigators was arranged.

It was specifically arranged that Dr. Ogura would get one of the Ames Research Center's Apnea Detector to test out in his clinic.

It was agreed by the participants that there would be no record of the meeting discussion.

During the year the Special Programs Branch worked with the Section on Epilepsy of the Applied Neurology Branch to continue the use and evaluation of the Epilepsy Abstracts Retrieval System.

During this fiscal year, three university departments of neurology were given access to the data base for eight months. The results tend to reinforce the results of previous tests indicating that researchers and clinicians save many valuable hours of library work and have a better information base for the conduct of their research and clinical practice.

The CVD abstracts were added to this data for test and demonstration purposes. The CVD data base while small was impressive in showing what an expanded data base could provide. This activity could be the forerunner of a much expanded system in which the data base encompasses the broad field of neurology and otolaryngology. This project is being maintained at a low level of activity until sufficient funds for the expanded system become available.

The Special Programs Branch is also responsible for the Special Foreign Currency (P.L. 480) Program of the NINDS. This will be described in a separate section.

Special Report
Foreign Currency Credit Award Programs
(P.L. 480)

The foreign currency credit award or P.L. 480 Program is a program of research supported by United States owned foreign currencies which are not convertible to dollars. It is authorized by the Agricultural Trade Development Systems Act, 1954, Public Law 480, subsection 104K, which permits the use of foreign currencies, "to conduct and support scientific activities overseas, including programs and projects of scientific cooperation between the United States and other countries such as coordinated research against diseases common to all mankind, that are unique to individual regions of the globe, and promote and support programs of medical scientific research."

At the present time funds are available in Burma, Guinea, Israel, Morocco, Pakistan, Poland, Tunisia, the UAR, Egypt, Yugoslavia, and India. This list is subject to changes and frequently does. Ceylon and Indonesia were on the list but have recently been withdrawn. There is some possibility of excess currencies being available in the Sudan. The research supported by P.L. 480 funds was originally primarily for medical and health research but the United States policy is being changed to allow many more U. S. Government agencies to use them. Therefore, the amount of money for medical research is being restricted.

At the present time, the Institute has 29 ongoing projects with a current annual dollar value of about $950,000 depending on the international money market. There is one project in Egypt, eight in India, three in Israel, six in Poland and eleven in Yugoslavia. In Israel two of the three projects will be terminated this year, and the last one next year. They are all dissimmilar. One study in Israel to elucidate the various neural endocrine factors regulating hypothalmic activity will terminate this year (06-008-1). Another problem is concerned with the central and autonomic control system modulating alimentary receptor activities and regulation of feeding behavior (01-015-1). The other study is a large program to understand the function of the nervous system by studying physiological and biochemical correlates of behavior. In addition to these studies it also is a development of a major research source, the International Brain Research Laboratory in Yugoslavia (02-003-1).

There are six projects which are basically neurochemical or metabolic chemistry studies, but they are generally unrelated. One is a study of the tolerance limiting role of metabolic events occurring in the brain at conditions of anoxia induced at different levels of hypothenia down to zero degrees centrigrade (02-042-1). There is a study of the role of neuroglia cells in the myelination and demyelination with emphasis on the enzyme chemistry of the processes. This of course relates to human demyelinating diseases such as multiple sclerosis (05-027-1). A somewhat similar study is a study of the glia of the nervous system and their function in diseases of the brain.

Particular attention will be given to the intracellular oxidation reduction enzymes (05-004-1). The subject of another study is the maintenance of the surface characteristics of membrane systems and the enzyme actions required to maintain these membrane systems. An initial approach will be the study of the action of sialidases on acid containing substances which occur in membrane systems (02-032-1). The control and regulation of protein synthesis in biological systems is the focus of another study (02-020-1). There is one project on molecular biology and biochemical studies of trachoma agents which is a holdover from the time when the NINDS had responsibility for eye research. This project is in Israel and will terminate this year (06-028-1).

There is one neuropathological study, just initiated, which is a study of a fine structure of the brains of rabbits with hereditary tremor. This tremor is similar to that seen in Parkinson's disease and this study may have some bearing on that disease (05-035-1).

There are six studies which relate to the neurological diseases of children. One of these is purely biochemical in nature; to synthesize the sphingolipids of the brain. This has been important because these substances have been used to elucidate the enzymatic defects of such diseases as Tay-Sachs disease (06-015-1). Another study which is both biochemical and clinical is to study the enzymatic processes involved in the sulfation and desulfation of glycosaminoglycans and cerebroside sulfate. These biochemical mechanisms have been studied in specific metabolic disorders of children. Using material from these patients the scientists working on this project have identified specific enzyme defects (01-024-1). There are two neuropathological studies relating to childhood. One is a study of the histogenesis of the forebrain in the human embryo using material from products of abortions (02-006-1). The other study is a neuropathological study of the anoxia damage of the immature nervous system studying the brains of newborns and infants who succumb to early anoxia (05-028-1). There are two studies that are related to the nutrition of the brain. One is the effect of protein malnutrition on the brain, metabolism, and development (01-041-1). The other is a general study of the nutritional disorders of the nervous system particularly those defects associated with malnutrition, the malabsorption syndrome, and peripheral neuropathies of uncertain etiology which occur in India (01-011-1).

There are two projects relating to trauma of the nervous system. One is a neuropathological study of closed brain injuries to determine the topographic distribution and histopathology of close head injuries (02-025-1). There is a study of the role of arterial venous malformations of the spinal cord which cause paraplegia and other myopathies of unspecified origin (01-099-1).

One project is a study of clinical angiographic and anatomic aspects of cerebrovascular disease in India (01-016-1).

There is one project which is an extensive multiple disciplinary program for the study of muscle disease in Poland (05-002-1).

There is one study on the biology and histochemistry of gliomas using tissue culture and immunochemical methods (05-013-1).

There is one project on the vestibular influences on normal and abnormal cerebral electric rhythms to see how these vestitular inputs contribute to the spread of cerebral electric rhythms and to correlate these with clinical problems (02-023-1).

In Yugoslavia the involvement of other U.S. agencies in the P.L. 480 program has resulted in a gross overplanning for the use of available funds. A joint U.S.-Yugoslavian committee has met and are trying to work out an extension of the program with contributions from both the United States and Yugoslavian sides. If this is successful then the program will continue, if not, available funds for the Yugoslavian activities will be used up by about fiscal year 1976. At present, we have 14 projects in Yugoslavia which have been approved, but may never be funded.

Each of the P.L. 480 projects must have a U.S. scientist to serve as a collaborating "Project Officer" who works with the foreign scientist in varying degrees. The role of the U.S. Project Officer varies from that of an active collaborative participating in the research to that of an informed sponsor. However, the project officer must each year review the expenditures and the progress report and advise the Institute whether the progress has been satisfactory. Of the 29 projects, nine have project officers who are in either the NINDS intramural or C&FR programs. The other 20 project officers are in universities throughout the country.

At the present time there are about 20 projects in various stages of development in addition to the approved, but unfunded projects in Yugoslavia. The agreement with Poland for use of P.L. 480 funds has been between the Polish Ministry of Health and DHEW, but in some of our new projects the scientists are in university laboratories. These university laboratories come under the Polish Ministry of Education so that to use P.L. 480 funds an agreement between the Ministry of Health and the Ministry of Education had to be worked out to allow the use of P.L. 480 funds to support research in the university laboratories. Such an agreement has been worked out, and it will considerably increase the number of scientists and laboratories which might be supported.

The expansion of the P.L. 480 programs is slow at times because of the changing political climates. The rebellion in East Pakistan with the establishment of Bandladesh wiped out several projects in Dacca. In countries like Guinea and Burma it is hard to find capable scientists with facilities to work out supportable projects.

It has always been difficult to find scientists who are capable of doing research even in countries that are fairly well developed. All the ex-holders of NIH International Fellowships who have trained in this country and returned to their native land and are working in the areas of interest to NINDS have been contacted to see if they can develop suitable projects. This

has met with a certain amount of success and many of our developing projects are from former fellowship holders.

The scientific content of the P.L. 480 research is complementary to other NINDS research and covers a wide area including both clinical and nonclinical subjects.

Three of the projects are concerned with naturally occurring neurotoxins. One is concerned with obtaining material of the poisonous animals such as snakes and others from the Middle East, North Africa, as far south as the Union of South Africa. Biochemical and physiological studies will be carried out on the purified toxins which will also be available to the investigators in the United States and antisera will be developed (03-006-1). Another similar study is being carried out in India, studying the poisonous snakes of India (01-040-1). The other project is on neurotoxins from plant materials, and is focusing on toxicity of B-diaminopropionic acid (ODPA) which is extracted from the seeds of Lathyrus sativus in India. This will be studied in connection with the development of human lathyrism (01-034-1).

There are five projects concerned with synaptic transmitters. One is to purify and characterize acetylcholinesterase to determine its molecular weight and other chemical characteristics (02-027-1). Another is to determine the role of the acetylcholinesterases in the acetylcholine system transmission conduction of excitation (02-008-1). There are two others which are somewhat related in the study of the metabolism release of five hydroxytryptamine and its function as a transmitter in the brain (02-015-1 and 02-041-1). These four are in Yugoslavia. Another project, in India, is a study of factors which alter the uptake and retention in various tissues of catecholamines and drugs which affect their action. The drugs studied are primarily the catacolamines and catacolamine depletors and other drugs which either mimic their effect or block it (01-109-1).

There are three projects which are essentially neurophysiological in nature.

Contract Narrative
Special Programs Branch, C&FR, NINDS
Fiscal Year 1973

The Brain Information Service at UCLA (NIH NINDS 70-2063)

Title: Operation of Specialized Information Center on Nonclinical Neurosciences

Contractor's Project Director: Michael Chase, Ph.D.

Current Annual Level: $410,000.00

Objectives: To operate, as a part of the National Institutes of Neurological Diseases and Stroke Neurological Information Network, a specialized information center for the nonclinical neurosciences to serve as a national focal point for the identification, collection, storage, retrieval, analysis, repackaging, and dissemination of information. The major thrust of this information center shall be information analysis products and services. The center will identify and locate all information items and services pertaining to its subject area. The center will use the identified and stored information to make available comprehensive information service primarily to the scientists supported by NINDS, secondarily to the remainder of the biomedical community throughout the country and, when possible, internationally. These services shall include: a) current awareness services; b) bibliographic services; c) translation of foreign language articles when essential; d) answering scientific questions relating to the subject area; e) promotion and preparation of reviews and other special articles for the general as well as specialized segments of the biomedical community, organizing and supporting workshops on specialized topics in the brain sciences and publishing their proceedings where necessary; f) furnish information about ongoing research; g) preparation of a publication series in the brain sciences.

Major Accomplishments: The Brain Information Service currently regularly sends to approximately 5600 scientists scientific information relevant to their needs. Approximately 6800 additional scientists received some other service or publication during the year. The current alerting bulletins produced are:

1. Sleep Bulletin and Sleep Review are monthly bulletins with 2071 subscribers. These bulletins have proved so useful that the Association for the Psychophysiological Study of Sleep has requested that all members have copies of the bulletin and have made subscription to them a requisite for membership in the association. As of January, 1973, a subscription charge of $15 per year was instituted.

2. Biogenic Amines and Neurotransmitters in the Nervous Systems - an annotated bibliography which appears semi-monthly and contains several one-page reviews or abstracts in depth. Distributed to 970 subscribers at a charge of $12 per year.

3. Neuroendocrine Control Mechanism: Hypothalmic-Pituitary-Gonadal System - an annotated semi-monthly with several one page reviews or abstracts in depth. Distributed without charge to 2500 subscribers.

4. Memory and Learning - Animal Studies - a monthly listing of published studies. Distributed without charge to 1668 subscribers, an increase of 85% over the end of FY72.

5. Memo of Current Books in the Brain Sciences - comes out monthly and is annotated. Distributed without charge to 2127 subscribers.

The Index to Current (EEG) Literature is prepared by the BIS as a quarterly supplement to the journal Electroencelphalography and Clinical Neurophysiology. The cost of printing and distribution is borne by the EEG Society.

In addition the Brain Information Service has developed two hard-bound series: Perspective in the Brain Sciences and Sleep Research. Two publications were developed this year for the Perspective in the Brain Sciences series: Volume 1, The Sleeping Brain in which an overview of the total field of sleep research is presented based on the symposia held at the First International Congress of the Association for the Psychophysiological Study of Sleep in Bruger, Belgium,and Volume 2, Operant Control of Brain Activity based on a conference held at the Kroc Foundation in Santa Barbara, California, attended by ten of the most prominent basic and clinically-orientd scientists in the field.

The Sleep Research series, Volume 1, 1972, contains expanded abstracts of the year's Association for the Psychophysiological Study of Sleep meeting, current claims of research findings and all pertinent bibliographic information for the year.

The Brain Information Service is placing more emphasis on the preparation of research review papers in specific areas of scientific endeavor. Manuscripts prepared thus far are Neuronal Modelling and Neural and Biochemical Circuitry Underlying States of Vigilance.

The Conference Report Series has been continued. These reports have been in great demand with as many as 3500 requests for a single report. Four reports produced this period are: (1) IV International Congress of Endocrinology - HGG Symposia, Washington, D. C., June 18-24, 1972. This is a 16-page report which covered the sessions on light and reproduction, brain

monoamines and the control of anterior pituitary function, brain control of the hypothalmus and pituitary and behavioral correlates of harmonal effects on the brain; (2) Neurobiological Aspects of Maturation and Aging, Downstate Medical Center, June 26-29, 1972. This 22-page report covered the sessions on anatomical and physiological correlates of maturation and aging, myelination, biochemical correlates of maturating and aging, and model systems. 3) Brain Research Institute Tenth Anniversary Symposia, UCLA, July 6-8, 1972. This is a 70-page conference report covering eight symposia and including an introduction by Dr. Theodore Bulloch entitled "The Brain is not a Horseradish: The Special Nature of Neuroscience." Symposia covered are electrophysiological processes in the epilepsies, neurobiology; evolutionary and developmental approaches, functioning brains mirrored in man-machine interactions, biochemistry of the synapse, what have we learned about learning, graded processes in the central nervous system, membrane organization and function in nervous tissue, and feedback actions of gonadal steroid hormones on brain, pituitary and behavior. (4) Second Annual Meeting of the Society for Neuroscience, Houston, October 8-11. Important lectures and symposia are covered in a 62-page report.

Significance to Biomedical Research: The Brain Information Service performs an essential service to the biomedical community by distilling and repackaging all new information items within its area of responsibility. An indication that this program is becoming central to the advancement of research in the basic brain sciences is the formal relationship being developed with the BIS by the Society for Neurosciences in which the Society looks to depend more and more on the BIS as a resource to the neuroscience community.

Proposed Course of the Contract: This program, as part of the Neurological Information Network, is under the surveillance of the Science Information Program Advisory Committee and its performance is reviewed annually.

Contract Narrative
Special Programs Branch, C&FR, NINDS
Fiscal Year 1973

The Johns Hopkins University (NIH-71-2281)

Title: Operation of an Information Center on Human Communications

Contractor's Project Director: John E. Bordley, M.D.

Current Annual Level: $337,600.00

Objectives: To operate as a part of the National Institute of Neurological Diseases and Stroke Information Center Network, a specialized information center on speech, hearing and human communication to serve as a national focal point for the identification, collection, storage, retrieval, analysis, repackaging, and dissemination of information. The major thrust of this information center shall be information analysis products and services. The center will identify and locate all information items and services pertaining to its subject area. In addition, the center will use the identified and stored information to provide comprehensive information service to the biomedical community throughout the country and, when possible, internationally. These services shall include: a) current awareness services; b) bibliographic services; c) translation of foreign language articles when essential; d) answering scientific questions relating to subject area; e) promotion and preparation of reviews and other special articles for the general as well as specialized segments of the biomedical community; f) furnish information about ongoing research and g) develop new methods of information analysis and dissemination.

Major Accomplishments: Current Citations on Communications Disorders is the major current awareness product of the Center, having a free circulation of 1,365. In December, 1972, this bulletin was divided and distributed in two parts: (1) Hearing and Balance and (2) Language, Speech, and Voice. These bulletins provide up-to-date coverage of articles, editorials, chapters of books, small monographs, unpublished papers, reports, etc. in the field of the communicative sciences. The audience includes scientists, clinicians, educators, administrators and students.

Biblioprofiles are comprehensive bibliographies on selected subjects that include a short state-of-the art digest by a scientist expert in the subject area. These products are available for a nominal charge that covers the cost of reproduction and mailing. The following is a list of Biblioprofiles that have been or are expected to be produced this year.

No. 3 Surgical Treatment of Deafness
No. 4 Homotransplantation: Part 1. Speech and Hearing Organs, Part 2. General
No. 5 Viral Infection and Hearing
No. 6 Auditory Physiology

No. 7 Rehabilitation of Language Disorders in Children
No. 8 Causes and Pathology of Deafness
No. 9 Speech Development

Prepared Bibliographies are produced periodically and are comprehensive bibliographies on selected topics. These had been available free but are now available for a nominal charge that covers reproduction and mailing. For example, a recent bibliography on Noise: Potential Danger to Man is available for $3.00 per copy. The production run was for 1000 copies. Other bibliographies produced or expected this year are: "Speech Production of the Deaf," "Rehabilitation of the Laryngectomized Patient," "Identification of Infant Hearing Impairment," "Geriatric Hearing Impariment," "Auditory Abilities and Academic Skills," "Deafness and Mental Retardation," "Language in Mentally Retarded Children," "Dyslexia," "Abnormal Language and Speech Development in Children," and "Cerebral Palsy and Communication Disorders."

Reviews produced as a result of special conferences and workshops are produced periodically. Reviews expected to be completed or worked on this year are "Neuroanatomy of the Auditory System" and "Vascular Disorders and Hearing Defects."

Critical Reviews are done occasionally. This year a whole journal issue of the Archives of Otolaryngology is used for eleven papers on the "Effect of Virus Infection on Hearing."

Handbook. The "Index Handbook of Ototoxic Agents" was produced this year. It provides a reference manual containing informative abstracts of the ototoxic drug literature, 1966-1971. Also created is a computerized data base accessible for searches of the ototoxic drug literature.

Special Products included an article in Military Medicine with a circulation of 9800 on "An Information Center's Place in Military Medicine." Also an "Announcement" was produced and distributed to users and others in the subject field to acquaint them with the products currently available from the Center.

Significance to Biomedical Research: The Information Center for Hearing, Speech and Disorders of Human Communication performs a valuable service to the biomedical community by distilling and repackaging all new information items covering the many disciplines in its area of responsibility. Special attention is given to the information needs of those whose programs are supported by NINDS funds.

Proposed Course of the Contract: This program, as part of the Neurological Information Network, is under the surveillance of the Science Information Program Advisory Committee and its performance is reviewed annually.

Contract Narrative
Special Programs Branch, C&FR, NINDS
Fiscal Year 1973

Clinical Neurology Information Center (NIH-NINDS-72-2300)

Title: The operation of a Clinical Neurology Information Center

Contractor's Project Director: Walter J. Friedlander, M.D.

Current Annual Level: $96,571.00

Objectives: To develop, as part of the NINDS information center network, a specialized Information Center on Clinical Neurology. This Center will be an international focal point for information relating to those diseases of interest to NINDS, especially information relating to diagnosis, treatment and prevention of diseases of the brain and central nervous system. The Center will produce reviews of various clinical problems of interest to the Government, bringing together the relevant clinical knowledge as it applies to the problem. These reviews may focus on an entire disease problem as a whole, or on any distinct part of a disease entity. The Center will identify sources of information relevant to clinical neurological problems, including indexing services, abstracting services, periodical journals, books, monographs, etc. In execution of this duty, the Center will explore the feasibility of answering questions about clinical neurological problems.

Major Accomplishments: In its first year of operation, the Center contracted with leading clinical specialists for a number of critical reviews. The manuscripts completed or expected this year are the following:

1) The Pharmacokinetic Basis of Meaningful Therapeutics in Medicine
2) Developmental Dyslexia: Neurological Aspects
3) Angiography of the Spinal Cord: 1973 State-of-the Art
4) Recent Concepts of Spinal Cord Injury
5) Minor Hemisphere Syndrome
6) Evaluation of Speech Therapy in Acquired Aphasia
7) Neurological Aspects of Hallucinogens
8) The Role of Neurotransmitters in Altering Seizure Susceptibility
9) Physiology of the Cerebrospinal Fluid (with particular reference to normotensive hydrocephalus)

Preliminary arrangements have been made for another series of critical reviews. The topics are as follows:

1) Effects of Malnutrition on the Maturing CNS
2) Clinical Evaluation of Neuropsychological Tests
3) Surgical Treatment of Aggressive and Violent Behavior
4) Evaluation of Physical Therapy in Fixed Neurological Disability
5) Chemotherapy of Brain Tumors

6) CNS Aging
7) Cranio-cervical Malformations
8) Evaluation of Serum Anticonvulsant Levels in the Treatment of Epilepsy

The Center's most innovative product and one that has been received enthusiastically by the approximately 1000 scientists who receive it is the Concise Clinical Neurology Review. This experimental publication emphasizes a one sentence (terse) abstract of each paper in a cluster of terse statements on a single topic. A number of these topics are covered in a single issue of the bulletin. The bibliographic citations are referenced by a number and appear together in a second part of the bulletin. Approximately 240 papers are selected for inclusion each month, based on a review of 692 serials. This publication seems to fulfill a need not otherwise met in neurology.

Another accomplishment was the publication and distribution of a Compilation of Material Available from Volunteer Health Organizations and from the Neurological Information Network.

Significance to Biomedical Research: This Information Center is visualized as a significant vehicle for bridging the gap between the researcher and the clinician. Full interaction with the other NINDS Scientific Center is assured.

Proposed Course of the Contract: This program, as part of the Neurological Information Network, is under the surveillance of the Science Information Program Advisory Committee and its performance is reviewed annually.

Contract Narrative
Special Programs Branch, C&FR, NINDS
Fiscal Year 1973

Cerebrovascular Disease Abstracting and Retrieval Service at the Mayo Foundation (PH 43-66-933)

Title: Cerebrovascular Disease Abstracting Service

Contractor's Project Director: Robert G. Siekert, M.D.

Current Annual Level: $31,800.00

Objectives: To provide a bibliographic alerting service of new literature on cerebrovascular disease by preparing abstracts of appropriate articles for publication in Stroke - A Journal of Cerebral Circulation.

Methods: The abstracts are from pertinent articles culled from a review of about 148 serial journals. Either the author's abstract is used or a new one is prepared if the author abstract is too long or otherwise unsuitable. They are then given to the editors of Stroke for incorporation in the journal.

Major Accomplishments: The journal Stroke carries in each of its bimonthly issues approximately 100 abstracts prepared under this contract.

Significance to Biomedical Research: For several years, these abstracts were offered to a small group of researchers on an informal basis. As more and more scientists heard about this service, the demand for the abstracts became so great that a way of general distribution was needed. The publication of these abstracts as a special section in the journal Stroke makes the service available to all those who would find it useful.

Proposed Course of the Contract: This program, as part of the Neurological Information Network, is under the surveillance of the Science Information Program Advisory Committee and its performance is reviewed annually.

Serial No. NDS(CF)-73 SP 2041
1. Special Programs Branch
2. Office of the Chief
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Evaluation of the Neurosurgical Biblio-Index

Previous Serial Number: None

Principal Investigator: Edgar A. Bering, Jr., M.D.

Other Investigators: None

Cooperating Units: Journal of Neurosurgery and the National Library of Medicine

Man Years:

Total:	0.15
Professional:	0.05
Other:	0.01

Project Description:

Objectives: The object of this project has been to determine the value of a recurring bibliography printed from the MEDLARS data file and especially designed to serve a particular group, in this case, the neurosurgeons. The Neurosurgical Biblio-Index is distributed with the Journal of Neurosurgery which has a circulation over 6000. Thus it includes many neurologists and nonclinical scientists as well as the neurosurgeons, so that this study provides data regarding the value of the Biblio-Index to scientists peripheral to the main interest of the bibliography.

Methods Employed: This study was carried out by a questionnaire method in two phases.

The first phase of the questionnaire primarily designed to identify user groups, whether they were neurosurgeons, neurologists, residents, nonclinical scientists, librarians or others. They were also identified as to whom among the clinicians were associated with medical schools and who were simply in private practice.

They were asked to rate the value of the Biblio-Index as to "valuable, very voluable, or of negligible value," and if they would pay $4.00 to $10.00, $10.00 to $15.00, or whether they would want it at all if there were a fee. They were asked if they would be willing to participate in further in-depth analysis.

Second phase questionnaire was considerably longer and sent to a sample from each of the six groups identified in the first phase. The respondents were asked how they used the Biblio-Index, how often for browsing, searching a specific subject (if so how often), whether they read it regularly, how it had changed their reading habits, what bibliographic sources they found most valuable, and whether its use made them more aware of new literature.

The study was not intended to evaluate the contents of the Biblio-Index as this has been designed by the Editorial Board of the Journal of Neurosurgery and was felt to be satisfactory. It was aimed at trying to get at the value to the users.

Major Findings: Eighty-six percent of the queried felt the Biblio-Index was either valuable or very valuable. Only a small number felt it was of negligible value. Some seventy-six percent were willing to pay up to ten dollars for it if it were put on sale.

It was found that such a bibliography was not of particular use to librarians, as they felt that it contained the same material as Index Medicus and they needed only the two tool (Index Medicus) to answer needs from many different groups. In libraries the Biblio-Index as filed as a supplement to the Journal of Neurosurgery and not put with the other index publications where people normally go for searches. Its value seemed to be the fact that it was on the user's desk or in his own library and was instantly available for searching.

The Biblio-Index did not seem to reduce the average number of journals that most people read regularly although about 20 percent of respondents did indicate some reduction. The mode of use was fairly uniform through all six groups of users (except librarians) which was surprising, particularly in the case of nonclinical researchers who seemed to find it very useful. The majority of people felt that it gave them better control of current literature as they were able to use the Biblio-Index both for browsing and for looking up special topics. Of those who answered the question, "Has it (the Biblio-Index) had an appreciable effect on their information gathering?" the only people who answered "no" were about 17 percent of those affiliated with medical schools, and this was true both with neurologists and neurosurgeons. All of the clinicians in private practice (both neurosurgeons and neurologists) said it appreciably helped their ability to gather information.

The survey of those who said the Biblio-Index was of negligible value were mostly librarians who felt Index Medicus satisfied their needs. The clinicians who felt that the Biblio-Index was of little value gave as their reasons that they did not do any writing or research and had no need for it, or that they had other people do their searching.

The NINDS, the Scientific Information Programs, and the National Library of Medicine were interested to find out the value of recurring bibliography because many of the products of the Neurological Information Network are in a sense recurring bibliographies for specialized areas, and the National Library of Medicine produces many recurring bibliographies in specialized areas. The conclusions that have been reached are that recurring bibliographies for individual retention and use are valuable to the specialists for whom they are designed and are also of value to scientists working in peripheral areas. Most people seem willing to pay a modest fee for them.

Based on the results of this survey the editors of the Journal of Neurosurgery and the American Association of Neurological Surgery were able to make the decision to increase the subscription price of the Journal to cover the production costs of the Biblio-Index, and to continue it as a quarterly supplement to the Journal.

Proposed Course of the Project: Final report is in draft form and being reviewed for distribution.

Honors and Awards: None

Publications: None

Serial No. NDS(CF)-73 SP 2042
1. Special Programs Branch
2. Office of the Chief
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Parkinson's Disease and Related Disorders

Previous Serial Number: None

Principal Investigator: Mr. Alfred Weissberg

Other Investigators: Edgar A. Bering, Jr., M.D.
Mrs. Lillian Kaplan

Cooperating Units: Bibliographic Services, National Library of Medicine

Man Years:

Total:	0.40
Professional:	0.20
Other:	0.20

Project Description:

Objectives: The monthly bulletin, Parkinson's Disease and Related Disorders: Citations from the Literature is a concise bibliography of papers from the current literature of interest to those in the subject field. It is sent to a user group of approximately 1000 located in more than 50 countries. This current awareness service is intended to help researchers and clinicians maintain knowledge of developments in the field of Parkinsonism.

Methods Employed: The bulletin is produced monthly by a special search of the newly produced tapes of the National Library of Medicine MEDLARS system. The NLM provides camera ready copy, composed by the computer and it is sent to the Government Printing Office for printing and distribution.

Significance to Biomedical Research: Current awareness bulletins help keep researchers and clinicians up-to-date on developments in their subject, saving them many hours of library work, reducing the likelihood of duplication of research, and providing new knowledge to the clinicians at an early date. The value of the

Parkinsonism bulletin to the neurological community is attested to by its enthusiastic acceptance.

Proposed Course of the Project: The mailing list for the bulletin is being revised and consideration is being given to instituting a nominal charge to all subscribers. An evaluation of its value to recipients is being carried out by a questionnaire method. The results of this should be available by the end of the year.

Honors and Awards: None

Publications: None

Serial No. NDS(CF)-73 SP 2043
1. Special Programs Branch
2. Office of the Chief
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Information Sources for the Neurosciences

Previous Serial Number: None

Principal Investigator: Mrs. Lillian S. Kaplan

Other Investigators: Edgar A. Bering, Jr., M.D.

Cooperating Units: Committee on Scientific Communication, Society for Neuroscience

Man Years:

Total:	0.35
Professional:	0.25
Other:	0.01

Project Description:

Objectives: The objective of this project is to produce a directory of the majority of sources of information relating to the general area of neuroscience. It will contain such things as core journals relating to the neuroscience, and a statement of their scope, what audiovisual aids exist for the neurosciences and where they are to be obtained, book series relating to neuroscience and special programs designed to provide information relevant to the neurosciences on a regular basis. Individual textbooks are not to be included. This will be a reference book for every scientist to have on his desk or in his laboratory which will assist him when he needs to have critical information relating to his research.

Methods Employed: The sources of the Brain Information Service at UCLA and the Information Center for Hearing and Speech and Disorders of Human Communication at Johns Hopkins University were reviewed; discussions and consultations were held with other information centers; the National Library of Medicine, members of the Council of Scientific and Technical Information, heads of other libraries, with scientists, with commercial publishers, considerable searching in libraries, and reviewing currently published serials.

Major Findings: A first draft has been written in five sections: 1) audiovisual aids, 2) bibliographies, abstracts, and index services, 3) information analysis centers, 4) journals, 5) directories, translations, and special organizations. This first draft of about 80 manuscript pages is being reviewed by a number of knowledgeable people for suggestions and omissions. After this a final draft will be prepared. The major difficulty has been in obtaining information about foreign specialized information centers or activities.

Proposed Course of the Project: After publication, the Society for Neuroscience wants to distribute it to all of its 1500 members, the British Research Council has asked for 1000 copies, and the American Neurochemistry Society has asked for 15 copies for review by their senior officers and are considering distributing it to all of its members. It will also be generally available to any other scientists probably for sale through GPO.

Honors and Awards: None

Publications: None

Serial No. NDS(CF)-73 SP 2044
1. Special Programs Branch
2. Office of the Chief
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1972 through June 30, 1973

Project Title: Cerebrovascular Disease Abstracts On-Line

Previous Serial Number: None

Principal Investigator: Mr. Alfred Weissberg

Other Investigators: Edgar A. Bering, Jr., M.D.

Cooperating Units: None

Man Years:

Total:	0.12
Professional:	0.04
Others:	0.08

Project Description:

Objectives: The Mayo Foundation prepares under contract approximately 600 abstracts on cerebrovascular disease for the NINDS. These abstracts are published in the journal Stroke. This project has as its objective making these abstracts available as part of an on-line neurology data base that can be searched with natural language queries. The first year's abstracts were made available on an on-line system during the fiscal year and the system was demonstrated to be operational and useful.

Significance to Biomedical Research: The availability of cerebrovascular disease abstracts on an on-line system provides biomedical workers in this field an opportunity to search quickly and easily through a data base of highly relevant material. When such services are available, they not only save valuable time for researchers and clinicians they can be instrumental in producing better research and patient care.

Proposed Course of the Project: The data base in cerebrovascular disease accumulated to date is very small and needs to be enlarged greatly to be of substantial value. The project is expected to continue building the on-line base at a rate of approximately 600 abstracts per year.

Honors and Awards: None

Publications: None

ANNUAL REPORT
Associate Director's Report
July 1, 1972 through June 30, 1973
Extramural Programs
National Institute of Neurological Diseases and Stroke

The Extramural Programs, NINDS, have been characterized in FY 1973 by (1) a continuation of the general decrease in the number of research grant awards; (2) an increase in program development in the stroke and the acute spinal cord injury research programs; (3) initiation of the phaseout of training programs; and (4) administrative reorganization.

Research Programs - As indicated in the report of the Research Grants Branch, the number of active research grants have decreased 40% in period FY 1968 to FY 1973. This is the result of a decrease in funds available for research grants and an increase in the average cost of each grant. In addition, the number of applications for NINDS research grant support continues to increase and now is beyond the number of applications reviewed prior to the removal of vision research from NINDS program responsibility. The reasons for this marked increase in requests for NINDS research grant support are difficult to document but probably are related to: the growing momentum of scientific interest in research on the brain, the nervous system and hearing, language and human communication; the recent development of research methodologies and technologies which permit research advances in these areas; the availability of a pool of highly skilled basic research and clinical research investigators; and a decrease in opportunity for research support from other organizations including other research support components of the NIH. It is safe to estimate from FY 1973 experience that in FY 1974, 50 percent of research grant applications will be recommended for approval and of those approved, 18-20 percent will be able to be funded; put another way, 1 out of 10 research grant applications reviewed competitively by the NINDS in FY 1974 will be funded.

Despite these serious limitations in the research grant program, during FY 1973 NINDS-EP continued to move ahead aggressively with the development of needed research support in selected targeted areas of Institute responsibility: stroke, acute spinal cord trauma and deafness. During FY 1973, 3 acute spinal cord injury research centers were activated; 3 others were recommended for approval but not funded. Also, of the stroke acute care research units (SACRU) recommended for approval, 10 were funded for activation in FY 1973. It is anticipated that an additional 25 applications for stroke acute care research units will be received in FY 1974. In FY 1974, pilot efforts for additional program development will be focused on the areas of nerve regeneration, epilepsy, multiple sclerosis and brain tumors. The area of research on brain and aggressive behavior is under intense review in FY 1973 by Institute staff and the National Advisory Council. Upon completion of a report on the status of research in this sensitive area, program plans will be developed for needed FY 1974 activities.

Training - In accordance with NIH instructions, the phaseout of NINDS training programs has been initiated as described in the report of the Training Grants and Awards Branch. Provisions have been made to provide stipend support for all previously committed NINDS supported trainees and fellows and for new trainees to whom documented institutional commitments were made prior to January 29, 1973. Support for training environments also is being provided in accordance with environmental needs as related to the number of NINDS-supported trainees and fellows. Four manpower evaluation contracts were active in FY 1973 in which the present manpower status and future needs are being evaluated in each of the Institute's clinical research areas; a fifth manpower evaluation contract in the area of the neurological and communicative basic sciences is being processed for activation in FY 1973. These manpower evaluation reports should provide the Institute with a more firm basis for manpower planning for the future.

Administrative Reorganization - Because of increased NIH emphases on targeted research, Extramural Programs has proposed the reorganization of its program professional staff to provide more adequately for coordination of grant activities in research areas of special emphases. The Training Grants and Awards Branch and the Research Grants Branch will be abolished; these will be replaced by a Regular Programs Branch and a Special Programs Branch--the former focusing on investigator-initiated research and the latter on targeted research and on manpower. It is anticipated that the changes proposed in the processing, grants management and administrative service units will be instituted in FY 1974. This reorganization will provide for increased delegation of responsibility to program and service staffs, organization along lines more clearly aligned with program areas and increased capabilities in administrative services. With increased administrative requirements, however, the problems of program coordination can be expected to increase.

ANNUAL REPORT
July 1, 1972 through June 30, 1973
Extramural Programs
Data Analysis and Reports
National Institute of Neurological Diseases and Stroke

The Data Analysis and Reports Unit has completed the conversion of its manual data files to a computerized information system. Data is available for Fiscal Year 1965 through Fiscal Year 1973 on all research grants, training grants, fellowship awards and indirect trainee appointments funded by the NINDS Extramural Programs. The development of new procedures and the revision of extablished methods of retrieval have continued throughout the year in an effort to make the system responsive to the Institute's needs for program planning and analysis, and requests for current operating information. The Unit has continued to update and incorporate information on Intramural Research projects and Collaborative and Field Research projects as furnished by these programs so that all Institute sponsored projects may be reported by the same Program Classification system.

The Unit, in addition to the regularly published data books and fiscal year summary books, prepared special reports for presentation at the Institute's three Fiscal Year 1973 Advisory Council meetings. The reports reviewed the major Extramural grant program trends during the last five years in the areas of both research and training. The Unit has also responded to requests from the Office of the Director, NINDS, the NINDS Information Office and the NINDS Financial Management Branch for special or detailed program information.

The Unit has had staffing problems during the year which have delayed operations in some areas, but as the fiscal year ends the Unit is again fully staffed.

ANNUAL REPORT
July 1, 1972 through June 30, 1973
Extramural Programs
Grants Management Section
National Institute of Neurological Diseases and Stroke

During the period of curtailed funding, investigators and grantee business officials are more prone to submit questionable budget requests to compensate for reductions in other areas which has resulted in our devoting a greater degree of concentration to each grant's review. The more difficult fiscal and budget problems necessitate more man hours for each grant review.

As of July 1, 1972, Rebudgeting of Funds within NIH Grants (NIH 5202) became effective. This policy provides for a designated grantee institution official to authorize grant rebudgeting instead of submitting their requests to the awarding unit. The areas where grantee institutions may rebudget are:

1. Domestic travel for any budget period in excess of $500 or 125% of the amount for domestic travel in the approval budget.

2. Individual items of equipment with acquisition costs of $1,000 or more, and total expenditure for equipment in any budget period in excess of $1,000 or 125% of the amount for equipment in the approved budget.

3. Patient Costs in excess of those originally approved.

4. Alterations and Renovations costs up to the lesser of $75,000 or 25% of the total direct costs (less exclusions).

Included in the above policy is the provision that in the opinion of the responsible grantee institution official, no procedure, policy or precedent clearly applies to a rebudgeting question; advice should be sought of the awarding unit. The delegation of this rebudgeting authority to the grantees, requires that the Grants Management Staff review each budget in greater depth to determine a realistic budget submission. Should the Program Staff determine that the grant includes aspects or other areas requiring greater monitoring, a footnote entitled

"Funds may not be rebudgeted in accordance with Policy Guide No. 19, (NIH 5202), without prior approval of this Institute"

is included on the award notice. This places the responsibility for rebudgeting decisions with the awarding institute instead of the grantee. The

Institute has utilized this footnote on a number of grants in the current fiscal year. The Program Project Grants are each reviewed with the Program Administrator for their decision to use the footnote.

In the Indirect Cost area, the monitoring of Grants Management's copy of the "Attachment To The Notice of Research Grant Awarded" has called our attention to a number of incorrect indirect cost awards and/or revisions that were not corrected during the fiscal year ended June 30, 1972: notwithstanding the assurance that all "Attachments To The Notice of Research Grant Awarded" would be serviced during the fiscal year, the lack of action on these incorrect indirect cost awards improperly reflect the Institute's balance of funds available at the end of the fiscal year. As result of this, we are monitoring the files more closely noting if there is any pending indirect cost awards or revisions of greater 90 days. Should any result, we call this immediately to the attention of the Indirect Cost Management Section.

In the past fiscal year the Neurological Program Project Review Committee and the Communicative Disorders Review Committee have increased grants management participation in their grant programs requiring an increase in the site visit participation and budget determinations.

The Grants Management Specialists have been attending a number of the larger study section meetings during the past year. This participation has contributed to a better understanding of the grants to be awarded. The Grants Management Specialists are being engaged more actively in their areas, such as site visits etc.

The Grants Technical Assistant added to our office has made a valuable contribution. The Grants Management Specialist have involved her in Interim Supplemental Awards, Award revisions resulting from Research Career Development Awards, and Reviewing Reports of Expenditures. In addition, she has participated in the indirect cost revisions to our monthly research award compilation. The assistance furnished by the Grants Technical Assistant has enabled our Grants Management Specialists to better apportion their time to other management areas requiring their attention.

The closeout of research and training grant files are proceeding slowly. A number of files have required more than one request of investigators and grantee business officials to obtain the necessary management reports. The Closeout Files are sent to the Data Analysis and Reports Section for final progress report determination. Response is subject to manpower and priority requirements of their office.

A new program of grants, Stroke Acute Care Research Units has been assigned to one Grants Management Specialist, thereby concentrating all efforts by one individual in our office. The Communicative Disorders Program Projects have also been assigned to another Grants Management Specialist, likewise concentrating on one area.

A new procedure, Equipment Accountability and Disposition, for each terminated project period is being satisfactorily reviewed by our staff;

Dr. Ray approving the research grant equipment authorization and Dr. Summers the training grant equipment authorization.

Annual Report
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Introduction

The brain is by far the most intricate, sensitive and versatile organ in the body. As a result, it has been the subject of extensive study and research for centuries. However, it has yielded only slowly to scientific exploration because of its complexity and because of its relative inaccessibility due to being enclosed in the skull and due to the blood-brain barrier which separates the brain metabolically in many respects from the rest of the body. Nevertheless, research is gradually bringing a greater understanding of how the 10 to 13 billion individual nerve cells in the brain, together with the additional billions comprising the nervous system, work together to make the human body an effective and coordinated living organism.

The economic burden of the neurological, sensory, and communicative disorders amounts to billions of dollars each year, with immeasurable human suffering. Although the human spirit can often adjust to the effects of physical disability, even the greatest courage may be broken by the devastating consequences of brain injury or disease which may continue or exacerbate during the remainder of a person's life.

More than 200 disorders are known to afflict the brain, sense organs, nervous system and neuromuscular apparatus, the most familiar of which are stroke, head and spinal cord injury, epilepsy, multiple sclerosis, parkinsonism, muscular dystrophy, cerebral palsy, aphasia, brain tumors, and otosclerosis. These disorders lead to paralysis, loss of speech, paraplegia and deafness, and are among the major causes of death and permanent disability in the United States.

The research grant programs of the National Institute of Neurological Diseases and Stroke are focussed on the identification, stimulation, and support of essential research problems aimed at the improved diagnosis, treatment, and prevention of disorders of the nervous system, the neuromuscular apparatus, the ear, and human communication. They include disorders of the young (cerebral palsy, epilepsy, learning disabilities) of adulthood (head and spinal cord injury, multiple sclerosis, brain tumors), and of the aged (stroke, parkinsonism, otosclerosis). The administrative instruments used to accomplish these purposes include research projects, research program projects, clinical research centers, outpatient clinical research projects, and specialized research centers.

Injuries to the spinal cord are occurring with increasing frequency. Once the spinal cord degenerates in the area of injury, paralysis always develops. In March 1971, the NANDS Council recommended approval of plans to support a limited number of Acute Spinal Cord Injury Clinical Research Centers. Each Center's plan would include investigation, development and evaluation of improved methods of emergency treatment, rapid transportation, diagnostic

techniques, medical-surgical repair, and the training of required professional, scientific and technical personnel. Operation of one of these Centers would be a major undertaking for even a large medical institution. Therefore, modest funds were provided on a competitive basis for feasibility studies at six institutions to enable them to determine if they could develop the requirements for a full scale center. Five of them applied for support for a full scale center, but only three could be supported with the funds available. It is expected that the fully developed Center will ultimately contribute important information on the prevention of degeneration of the spinal cord and on the restoration of spinal cord function. Close liaison with other government and non-government agencies with responsibilities in this area is being maintained.

The importance of improved stroke acute care is obvious since 70 to 80 percent of mortality occurs in the first ten days. Also, the consequences of not recognizing a progression or extension of the infarct may be catastrophic. On the basis of an extended review of this problem by the Joint Council Subcommittee on Cerebrovascular Disease, NHLI-NINDS the Director of NINDS requested, and the NANDS Council approved the organization of an NINDS Commission on Stroke. On March 30, 1973, the Commission made its final report entitled, "A Blueprint for National Action Against Stroke," after nearly a year of study, discussion, and analysis. The 97 page report contains some 25 specific recommendations for a national program on stroke, the third leading cause of death in the United States and the Nation's major cause of long-term disability.

In June 1972 in the face of what appeared to be a favorable fiscal situation and with knowledge of an urgent need, the NANDS Council recommended the establishment of a program of Stroke Acute Care Research Units (SACRU). Grants not to exceed $75,000 per year would be awarded to develop institutional focal points for (1) increased attention to the development or improvement of methods of prevention, diagnosis and treatment of the acute stroke; (2) development and evaluation of treatment methods and, (3) related training of professional and scientific personnel in this area. Twenty-six SACRU applications were received and 15 were recommended for approval by the initial review committee and by the Council in March 1973. In view of the limited resources at that time, however, the Council felt that only 10 SACRU could be financed this year without crippling other equally urgent programs. It is anticipated that the SACRU program will ultimately be a broad one to improve the prevention, diagnosis and treatment of the acute stroke episode at many medical institutions.

The number of applications received and, until this year, the number of grants awarded have been steadily increasing. The following Table shows the number of grants awarded and the total amounts of funds expended (in millions) each year for the past six years.

	FY '68	FY '69	FY '70	FY '71	FY '72	FY '73
Number	1,760	1,790	1,267	1,256	1,280	1,056
Dollars	$ 65.1	$ 67.8	$ 48.8	$ 53.6	$ 64.2	$ 62.4

The largest change was in FY '70 when all research on vision was removed to form the National Eye Institute. Since then the number of projects supported has been maintained at about the same level despite a marked increase in the cost of doing research. This was made possible in part by modest increases in the amounts of funds available, but primarily by the fact that from FY '70 through FY '72 every research grant, competing and committed, was negotiated downward from 6 to 15 percent. In FY '73 NIH decided that grants would no longer be reduced for fiscal reasons. Also, the total amount of funds available was reduced.

As shown in the following Table, the number of applications for each Council meeting is steadily increasing, amounting to an increase of 40 percent since June 1970.

	June '70	June '71	June '72	June '73
Number	339	352	398	467

This continued and dramatic increase in the number of requests is attributed primarily to the effectiveness of the research training programs of the Institute in which the output of fully trained investigators has only in recent years reached its full potential and which are now being phased out due to lack of funds.

Until recently the Research Grants Branch has been fortunate in maintaining a stable and experience professional staff. Last year three people left (a reduction of 37 percent) only one of whom has been replaced. With the increased number of proposals and the increased work as a result of funding so few grants, the staff has hardly been able to cope with the load. It is expected that this situation will be resolved during the coming year with the recruitment of an additional person and the assimilation of some research training grants staff.

The numbers of grants and the amounts of funds in the various disorder categories are shown in Appendix A.

FUNDAMENTAL RESEARCH IN THE NEUROLOGICAL SCIENCES

Introduction

The great importance of fundamental studies on neurological, sensory, and communicative components is frequently overlooked because they may have little or no immediate clinical application. However, the history of science shows clearly that improvements in the treatment of disorders are largely dependent on sound basic research.

It is generally accepted that the membrane around a nerve fiber plays an essential role in the transmission of the nerve impulse along the fiber. However, relatively little is known about the structure and mechanism of action of the nerve membrane or of living membranes in general. There is no doubt that research on membrane structure will contribute important information about how the nerve impulse is transmitted along the nerve fiber. There is a substance known as the nerve growth factor, the exact composition and source of which is still uncertain, which has a remarkable stimulatory effect on the growth of nerve fibers. The ultimate importance of the factor is still to be determined, but it surely will have important functions in nerve regeneration and it may well turn out to be useful in healing lesions and restoring function in the brain and spinal cord.

There are several anti-convulsant drugs that are relatively effective in controlling seizures in a good proportion of epileptic patients. Some of these drugs are quite unrelated chemically and there is no explanation why they are effective and why closely similar compounds are entirely ineffective. It has been suggested that the efficacy of a drug may depend upon a highly specific molecular structure rather than on the chemical composition of the molecule. Therefore, studies on molecular structure are now highly important and involve such sophisticated techniques as x-ray crystallography, electron paramagnetic resonance spectroscopy, and nuclear magnetic resonance spectroscopy. On the basis of such highly specialized information on the atomic interrelationships and the electron activity in the effective molecules, it will be possible to find or synthesize other molecules that are more effective in the treatment of disease. It may also lead to an understanding of how the present drugs work, which often is completely unknown now.

One of the great unknowns in neurophysiology has been the exact mechanism by which the nerve impulse is transmitted across the synapse from one nerve fiber to the next or from a nerve to a muscle or other end organ. In 1904 it was suggested that some nerves accomplished this by the liberation of epinephrine. In 1914 another investigator suggested that acetylcholine was the synaptic transmitter in some cases. These postulates were finally proven in 1921 by the classical experiment in which two frog hearts were connected by a small glass tube. When the beat of one heart was slowed by stimulating the vagus nerve, the beat of the other heart also slowed a few seconds later, demonstrating conclusively that some chemical was produced by the nerve in the first heart which was carried in the perfusion fluid and produced a similar effect in the completely separate second heart. It is now known that a number of compounds (acetylcholine, noradrenaline, dopamine, 5-hydroxytryptamine, gamma-aminobutyric acid, glutamic acid, glycine) may act

as synaptic transmitters. However, the mechanism by which the transmitter may be produced and act very rapidly (many times per second) and how the receptor nerve receives the stimulations is still largely unknown. As mentioned at the beginning of the Report, there are 10 to 13 billion nerve cells in the brain alone. Each then connects with from two to several other nerve cells. Therefore, the number of synapses is almost infinite and research on synaptic transmission is of the utmost importance. Also, this is a good example of why the application of basic research must be a time consuming process. The first evidence of the chemical transmission across synapses was obtained more than 65 years ago. However, the development of the area, even to its present unresolved state, had to await the discovery of highly sophisticated techniques such as electron microscopy and methods of chemical analysis sensitive enough to detect literally only a few molecules of a specific compound.

Nerve Growth and Regeneration

The clinical importance of being able to induce nerves to regenerate and resume their normal function is obvious. It would not only permit restoration of damaged spinal cord and peripheral nerve function, but it might ultimately allow reconstruction of damaged portions of the brain.

In general there are two approaches to this problem. One involves a study of factors such as temperature, drugs, etc. which affect nerve regeneration in spinal cord injured patients or in experimental animals in which a carefully controlled type and degree of nerve or spinal cord injury has been produced. The second general approach is the growth of nerve cells in tissue culture. This method has the disadvantage that the cells are in an abnormal situation, but it has the advantage that experiments can be more readily controlled and analyzed than in patients and animals.

Of the many things that affect nerve regeneration, the nerve growth factor (NGF) deserves special mention. It is of interest to note that its existence was first demonstrated in 1948 in a study of cancer in which mouse sarcoma 180 was being transplanted into chick embryos. It was noted that the cancer transplant became profusely innervated and, subsequently by a different investigator, that nerve growth was stimulated remote from the neoplastic tissue. It was established that the growth was produced by a diffusible substance more recently shown to be a relatively small, basic protein. Mouse submaxillary glands and snake venom have been shown to be the most potent sources of NGF. It effectively stimulates the production of nerve fibers from both sensory and sympathetic ganglia in culture and from sympathetic ganglia in vivo. In addition, the treatment of infant organisms with antibodies to pure NGF abolishes the growth of sympathetic nerves and ultimately causes complete destruction of the tissue. Thus, it appears that NGF is highly specific for nerve tissue. Some details of research in this area are described later.

Nerve fiber regeneration can now be stimulated by various means. However, the extremely difficult problem of synapse formation and the functional connection of a regenerated nerve fiber to another nerve or to a muscle still remains. Despite striking progress in fundamental studies of nerve growth and regeneration, restoration of function in the spinal cord injured certainly is not an immediate prospect.

Results of research on nerve regeneration were very discouraging until one group of investigators in 1957 reported axon growth through a Millipore sheath across a gap created by complete transection of a peripheral nerve. It appeared that the sheath protected the field of regeneration from invasion by extraneural scar tissue while creating a milieu which was favorable for axon migration. These results led to early attempts to develop methods to surgically repair severed cat spinal cords. These studies produced an excellent pattern of axon migration across a 4 mm gap in a completely transected spinal cord. Also, the regenerated axons conducted evoked action potentials up to 3.5 cm on each side of the pre-existing gap. However, no bioelectric evidence could ever be found that the regenerated axons made synaptic connections with other nerves. There was never any re-establishment of voluntary motor function.

In a different approach, nerve transection was produced by low temperatures (-20° to -30° C) around the spinal cord. This produced a 12 mm gap in which there was complete liquifaction of all neural tissue. In 14 days the gap was bridged by many thousands of myelinated axons. The axons were myelinated, some with peripheral type myelin as indicated by the presence of nodes, others with myelin of a central nervous system type. The usual Schwann cells and glial cells were present. However, repeated examinations of long-surviving preparations of this type failed to show any evidence of restoration of motor or sensory function caudal to the area of injury.

Other approaches are being utilized in the treatment of spinal cord injury, many of which are directed toward the prompt prevention or elimination of edema, toxic metabolic products and similar considerations which may cause secondary damage more serious than the original injury. With regard to nerve regeneration, however, the problem does not seem to be primarily one of simple nerve growth. The basic question is how does the regenerated nerve become functional? How can it be made to develop a normal synapse? What determines which nerves another nerve should form a synapse with? Some of these questions are touched on elsewhere in this report.

One investigator has approached the problem of nerve growth by efforts to (1) refine and extend available techniques for the dissociation, fractionation and culture of neural tissues; (2) investigate the possible role of non-neuronal cells in supporting neuronal survival in vitro and affecting their reaggregation patterns, and (3) relate the results of these studies to the effects of the nerve growth factor (NGF). The cells used were from the dorsal root ganglia of the newborn mouse (mDRGs) or the chick embryo (cDRGs).

Improvements in cell separation techniques previously used with cDRGs and sympathetic ganglia were successfully applied to mDRGs, yielding about 6,000 neurons and an equal number of non-neuronal cells per ganglion. When seeded over collagen-coated surfaces, neurons and non-neurons attached to the surface with different time courses, thus providing a method for bulk separation of the two main cell classes of the ganglia. The earlier attaching, non-neuronal elements (Schwann cells, fibroblasts, numerous less readily classifiable, irregularly-shaped cells) grew rapidly and could be propagated in culture through several passages. However, passed cells had a much more uniform, epithelioid appearance. The neuronal fraction, while

freed from readily attachable non-neurons, was not pure. Many of the neurons remained studded with tightly associated satellite cells. When re-seeded by themselves into collagen-coated dishes, the purified neurons failed to attach, grow fibers and survive in significant numbers even in the presence of NGF. They would do so, however, if re-seeded on their own original non-neuronal partners.

A quantitative analysis of the cultures of mixed mDRG cells revealed that attachment fiber production and survival of the neurons were markedly dependent on the availability of exogenous NGF in the culture medium which induced a 5-fold increase.

Attachment, fiber production and survival of mDRG neurons were also greatly enhanced when the dissociates were seeded over pre-attached non-neuronal ganglionic cultures in the absence of NGF. A moderate non-neuronal supplementation was adequate to mimic the NGF effect only over the early stages of the culture, but a more extensive supplementation fully substituted for the presence of NGF and, in fact, improved neuronal performance over that elicited by NGF. In all cases it was possible to confirm a supportive role of the homologous non-neurons on the culture behavior of the neurons and the ability of the non-neurons to substitute for the otherwise required presence of NGF in the culture medium.

This work places the NGF phenomenon in a new and different light by its overlap with the effectiveness of non-neuronal elements. The question is whether NGF may not mimic a non-neuronal physical or humoral action, facilitate an otherwise insufficient neuron/non-neuron interaction , or even act directly on the non-neurons while only indirectly affecting its neuronal elements. Perhaps most important, if the non-neuronal activity can be shown to come from glial cells, an experimental in vitro system would become avialable for the study of glial mediation in physiological or pathological events effecting the nervous system.

It is essential to know as much as possible about the composition and structure of the NGF molecule in order to know how it works. One laboratory has completed the entire amino acid sequence of the molecule and has gone on to try to identify the parts of the molecule that are essential for its activity. Somewhat surprisingly the structure and certain functional aspects are very similar to those of insulin.

Using ion-exchange media, the NGF molecule was separated into four parts. The first two parts seemed to lack the C-terminal arginyl residue. Treatment of native NGF with carboxypeptidase B also was shown to remove this residue. In neither case did the absence of this amino acid residue affect the biological activity of the molecule. Similar studies on tyrosine showed that it was not of primary importance in receptor binding in either NGF or insulin.

A variety of reagents were used to probe the location and functional importance of three tryptophan residues in the NGF molecule. A comparison of the residues in the corresponding positions in insulin indicated a close parallel which is consistent with the hypothesis that at least portions of the

two molecules possess similar structures. Bioassays indicated that all three tryptophan residues could be modified without loss of activity. Thus, these groups are not functioning as active sites of the molecule.

NGF labelled with ^{125}I is fully active. In addition to its value in radioimmune assays, it has been tested in a number of systems for its capacity to bind to responsive cells. Thus far chick sensory (dorsal root), chick sympathetic (superior cervical) and two responsive human neuroblastoma cell lines have been shown to bind NGF. Preliminary calculations show about 5,000 - 10,000 binding sites per cell in the sensory ganglia.

Studies of NGF from the venom of the cobra and the rattle snake indicate that the two are similar, that they are composed of only a single chain of amino acids, and that they have a molecular weight of about 20,000. These results suggest that the higher molecular weight forms of NGF isolated from mouse submandibular glands are not found in snake venom. However, snake venom NGF appeared to be just as active as mouse NGF on a weight basis. Thus, there may be at least two forms of NGF. This may be an advantage in determining the active points in the molecule, the receptor points or binding sites on the nerve cell, and ultimately putting the NGF to some clinical use.

The Synapse and Neuromuscular Junction

Brain function is remarkably stable, yet the brain can address itself to major changes. The brain adjusts to new experiences, but it also replays old ones with exquisite reliability. The coexistence of brain stability and plasticity is paradoxical.

Synaptic events probably underlie both the stability and plasticity of brain function. Alterations in the transfer of information by the synapses is achieved at two levels (1) the number of active synapses changes by making or breaking contacts; and (2) existing synapses are modified in such a way as to change the postsynaptic response. The central and critical question in synaptology is to determine the relevant and limiting molecular events guiding synaptic ability and change.

One laboratory has made a detailed biochemical and electron microscopic study of subcellular synaptic fractions including the synaptic complex (SC), the synaptic plasma membranes (SPM) and the postsynaptic density (PSD), a thickening subjacent to the plasma membrane on the postsynaptic side. It was found that proteolytic enzymes digested the PSDs, that the synaptic complex did not require protein simply for the structural integrity of non-protein components, but that the proteins themselves are functionally very important in synaptic structures. Electron microscope studies indicated that the PSD consists of a series of small (60 Å) granules interlaced in a fiberous-like matrix and that a set of bristles was attached to the postsynaptic plasma membrane exactly external to the PSD. Using labelled compounds, it was found that active binding sites were restricted to the bristles extending from the postsynaptic plasma membrane.

On the basis of this work, it is now possible to determine the adhesive forces or molecules in the cleft linking the pre- and postsynaptic plasma

memuranes. There are two models to describe the synaptic connection. One is that single molecules span the cleft linking pre- and postsynaptic membranes while another maintains that polyionic bonds are involved. These data on synaptic complexes provide basic molecular information about the characteristics of the immediate site of synaptic interaction. It will lead to information about what a synaptic complex is and how nerves are connected via this structure.

A study of the neuromuscular synapse has been approached by one investigator by innervating skeletal muscle with one of several types of nerves. Frog sartorius or pectoris muscles were transplanted to the lymph space under the skin. Into this space one or more of the following nerves were implanted: gastric vagus (parasympathetic preganglionic), splancnic (preganglionic sympathetic), posterior slpancnic (postganglionic sympathetic), skin sensory nerves. or the fourth or fifth spinal nerve (thoracic somatic motor nerve).

Skin sensory and postganglionic sympathetic nerves show no evidence of innervating skeletal muscle or of maintaining it in any way. The muscle atrophies and develops acetylcholine (ACh) sensitivity as if it were totally denervated. Preganglionic autonomic nerves (gastric, splancnic) are quite capable of forming functional synapses. However the weight of the muscle dropped to about 50% in six months as conpared to 20-25% in a denervated muscle. With somatic innervation, the drop was only to 60-80% of the original weight

In the case of vagal innervation, the number of muscle fibers decreases, but innervated fibers appear as healthy and large as control fibers. With splancnic innervation, the number of fibers decreases slightly, and the remaining fibers all look equally atrophied, despite innervation. It appears, therefore, that the gastric vagus nerve is better able than the splancnic to maintain healthy muscle fibers.

In most cases, not all of the muscle fibers become re-innervated by the autonomic preganglionic nerves. Vagus-innervated muscles could produce only 44% of the tension, and contained only about 50% of the number of fibers as compared to controls. Splancnic innervation was usually less successful with only 10-20% of the tension even though the number of fibers was not greatly reduced. Hence, either a portion of the fibers remain uninnervated, or the synapses are not sufficiently effective to produce stimulation in a large fraction of the fibers.

There are distinct differences in synaptic pharmacology between somatic motor nerve and autonomic nerve reinnervated muscle fibers. In the case of the latter there is no evidence, physiologically or histologically, of acetylcholinesterase (AChe) at the new junctions. Apparently the autonomic preganglionic nerves are incapable of inducting its synthesis or concentration at their synapses. Denervation leads to generalized ACh sensitivity on transplanted muscle just as does on other denervated muscle. Reinnervation by somatic motor nerves, the vagus or the splancnic all lead to a similar normalization of this condition. That is, the generalized sensitivity to ACh largely disappears and is restricted to the site of the new synapses.

Electron microscope studies of the neuromuscular junction of crayfish in another laboratory showed that two types of axon terminals could be

distinguished on the basis of structural differences in the synaptic vesicles. The excitatory terminals contained spherical vesicles about 500 Å in diameter. The other type had smaller, more pleomorphic vesicles. They were identified as inhibitory on the basis that they were sometimes presynaptic to the excitatory terminals, a morphological correlate of the presynaptic inhibition known to occur in these preparations.

Several examples of neuromuscular junctions have been reconstructed from electron microscope serial sections. The two types of axon terminals, as described above, both vary considerably in their physical relationship to the muscle fibers. Some make contact at only one point, others end in long "en passage" synapses. In a few cases, the axon terminals laid in deep clefts of the muscle fiber.

Repeated attempts to innervate skeletal muscle with autonomic nerves in the frog have not been very successful. Although there is some evidence that actual innervation occurs, this has been very difficult to confirm with electron microscopy because of the problem of finding the very fine tips of the regenerating autonomic axons and seeing their structural relationship to the muscle fibers.

Neurotransmitters

The goal of one laboratory is to elucidate the molecular events underlying chemical synaptic transmission using purified snake venom, a protein toxin, which strongly inhibits specific aspects of neuromuscular transmission. These macromolecules presumably bind to an extracellular site important in some aspect of synaptic function and, by blocking this site, lead to synaptic failure. Which aspect of synaptic function is blocked by a given toxin can often be determined and the biochemical nature of the binding site may be identifiable using radioactive toxin as an assay. Thus, using snake venom toxins as a probe, it should be possible to identify and describe biochemically the extracellular molecules involved in a known aspect of synaptic transmission. Such information would be invaluable, not only for understanding synaptic mechanisms, but also as probes for the measurement of plasticity in the nervous system.

One of the snake venom derivatives (alpha-bungarotoxin) specifically inhibits the acetylcholine (ACh) receptor, a postsynaptic element of neuromuscular junctions. This ability is now being used to study the neural regulation of the receptor and of acetylcholine esterase (AChe) in chicken muscle. This is of special importance since there is evidence that chicken dystrophic muscle has the same high AChe levels as denervated muscle, but the same receptor level as normal muscle.

When chicken muscle was denervated, there was an increase in the number of acetylcholine receptors in 3-4 days and an increase in AChe in 4-5 days. In 10 days the AChe level in a denervated muscle was 20 times higher than in the control muscle. This technique is being used to study a closely related strain of chickens which carries muscular dystrophy as a recessive gene. In this strain, the activity of AChe is about 20 times higher than normal, whereas the acetylcholine receptor level is normal. In addition to attacking the fundamental

problems presumably involved in muscular dystrophy, this study will resolve the current controversy over the relative importance of muscle activity or trophic factor release in the regulation by the nerve of the number of postsynaptic acetylcholine receptors.

Another investigator is studying the neural control of postsynaptic muscle proteins which play a direct role in synaptic transmission. Using organ culture techniques, attempts were made to see (1) whether changes in acetylcholinesterase (AChe) activity and acetylcholine (ACh) receptor distribution which occur after denervation could be duplicated in vitro; and (2) to see whether the changes which occur in these proteins after denervation are due to the loss of electrical and/or contractile activity of the muscle or to the loss of a more specific neural influence.

Using mammalian muscle, it was demonstrated by electrophysiological methods and with labelled alpha-bungarotoxin that muscles cultured for 5 days become sensitive to ACh over their entire surface. Such muscles bind toxin at non-endplate regions and individual fibers have sensitivities to ACh comparable to that in denervated muscle. Also, AChe activity decreases to nearly half of the activity of innervated muscle. Experiments are now in progress to attempt to stimulate muscle fibers in culture directly to see if changes in AChe activity and the ACh receptor distribution can be prevented or reversed by direct stimulation and muscle contraction.

The question of whether changes produced in muscle by denervation occur simply because of the loss of muscular activity was also studied in vivo on innervated muscles in which a chronic block of neuromuscular transmission was produced. Rats were maintained on a respirator for three days with continuous infusion of curare to block neuromuscular transmission. The distribution of ACh receptors was then determined by the binding of labelled alpha-bungarotoxin. It was found that regions of the muscle without endplates bound nearly 50 times as much toxin as control muscle. Thus, muscles in these animals behaved as though they were denervated. These results support the idea that increased ACh sensitivity in muscle is related to reduced muscle activity. Further studies are underway to determine unequivocally whether any specific neural influence is involved.

One laboratory has been so successful in separating and concentrating various subcellular synaptosomal fractions such as the synaptic complex, the synaptic plasma membranes and the postsynaptic density, that it is now possible to use these fractions to study transmitter release in vitro. The synaptosomes were pre-loaded with radioactive GABA, which is probably a major inhibitory transmitter in the brain, and then perfused with various solutions to alter the functional state of the synaptic endings.

It was found that synaptosomes depolarized by K were readily induced to release GABA by pulsing with Ca. The release was detectable in 30 seconds and maximal in 60 seconds. Because autoradiographic analysis had shown that synaptosomes are by far the predominant structure accumulating GABA in brain synaptosomal fractions, the secretion cannot be originating from a minor contaminant. It was also shown that release of GABA was blocked by Mg, Mn and other divalent ions known to block stimulus-secretion coupling in other

systems.

These data provided several new findings. The secretion occurs rapidly. It is blocked by Mg as required for a neurosecretory process. The signals are 10-20 fold larger than any reported by other investigators.

This system should provide a means to explore the mechanism of stimulus-secretion coupling with unprecedented resolution. It should be useful as an assay to determine the rate of CNS neurosecretion in various pathological conditions and after drug treatments. It may be possible, for example, to study neurosecretion in human biopsy samples and to evaluate its functional state associated with various disorders. It should also be possible to directly study the effects of drugs on neurosecretion and to explore potentially useful new drugs.

These few examples of research on nerve growth and regeneration, the synapse and neuromuscular junction, and neurctransmitters represent at best only a limited indication of the extreme importance of these areas and the amount of work going on in them. As suggested previously, an understanding of these fundamental problems would do more than anything else toward developing improved and effective treatment for many neurological disorders.

CONVULSIVE DISORDERS

Epilepsy is a disease characterized by one more of the following symptoms: (1) paroxysmally recurring impairment or loss of consciousness; (2) involuntary excess or cessation of muscle movements; (3) psychic or sensory disturbances; (4) perturbation of the autonomic nervous system. On the basis of origin, duration of seizures, clinical and electroencephalographic evidence, epilepsy is classified into four subdivisions.

In epilepsy known as "Grand Mal," in more modern terminology, "major motor seizures," the attack is most violent and convulsions last longer. The afflicted person may bite his tongue and invariably loses control of his faculties. If the area or areas of the brain can be located from which these abnormal electrical discharges emanate, the affliction is termed "Focal" or Jacksonian epilepsy. In "Petit Mal," a disorder of the young, the seizures are of short duration, and attacks occur more frequently. Seizures are characterized by myoclonic jerks and they generally start in the extremities and/or in one corner of the mouth. The affected part trembles violently. The trembling movement moves upwards. It ends up in a minor convulsion or the individual loses consciousness in the same way he does in grand mal. "Psychomotor" epilepsy is caused by discharging neurons which exert their influence upon mental processes as well as upon muscle movements. A patient exhibits adversive or torsion movements, dreadful fear, flinging of arms aimlessly, smacking of lips, and other incoherent physical and mental behaviors. The last type is called "Autonomic" epilepsy. A person suffering from it experiences a sharp, acute, sudden pain in the stomach. Hypertension, perspiration and other viceral disorders are the common symptoms.

Epilepsy may be caused by several factors, such as brain damage, presence of a scar caused by a wound or an injury, drugs, congenital malformation, nutritional deficiencies, metabolic abnormalities, fever (in infants), infectious diseases (encephalities, meningitis), brain tumors and abscesses.

A number of animal preparation displaying natural or induced epileptiform activity have been described as tools for the study of experimental epilepsy. For example, epileptiform activity has been induced by electroconvulsive shock; by systemic administration of convulsant drugs alone or in conjunction with brain lesions; by localized electrical stimulation of the brain; or by the creation of epileptic foci by direct administration to brain tissue of chronic irritants such as penicillin, cobalt, ethyl chloride, and alumina cream. "Natural" epileptiform activity has been triggered by auditory stimulation in rats, mice and rabbits and by intermittent photic stimulation in Senegalese baboons. The effects of various antiepileptic agents have been examined at one time or another in most of these preparations, and nearly all have been suggested as possible screening techniques for potential antiepileptic agents.

Methods based on electrically-induced seizures include generalized stimulation of the intact animal to produce either threshold (clonic) seizures or maximal (flexor-extensor) seizures, and local stimulation of selected areas of the brain or spinal cord. It is believed that the development of a well-studied reproducible model of status epilepticus that avoids the use of analeptic drugs will bring about a significant improvement in our understanding

and perhaps the treatment of status epilepticus which remains the leading cause of death from epilepsy.

A study is being conducted to investigate the nature and biological significance of ribosomal and other biochemical changes induced by electroconvulsive shocks (ECS) in the brain and the possible relationship of these modifications to brain damage resulting from multiple seizures. There have been many studies of cerebral protein synthesis in epilepsy. Several of them concerned with short term changes that might explain the amnesic effects of seizures. Although the initial studies produced conflicting results, the more recent attempts have developed a common consensus that brain protein synthesis is inhibited during convulsive seizures. Alterations of brain protein synthesis in epilepsy may have clinical applications: for example, it may mediate the impairment of brain development by seizures in newborn; its role in post-seizure amnesia has attracted wide interest; and the similitude in threshold and reversibility of self-sustaining status epilepticus and breakdown of brain polysomes suggests somelink between these two phenomena. Also, the inhibition of protein synthesis and breakdown of brain polysomes by repetitive cerebral seizures in the absence of convulsions raises the question of the role of protein metabolism in the brain damage that complicates conditions characterized by frequent or continuous electrical paroxysms without full tonic-clonic crises (e.g. petit mal status, West's syndrome). Administration of 50-100 consecutive electroconvulsive shocks to paralyzed, O_2-ventilated cats or rats resulted in the appearance of self-sustaining seizures that could continue for hours after the end of shocking. Using this model, a study of ribosomal changes in status epilepticus indicated no change in total ribosomal RNA or in the proportion of membrane-bound ribosomes after 100 ECS in paralyzed O_2-ventilated rats.

Ribosomal profiles showed a breakdown of polysomes, and an increase in ribosomal dimers in status epilepticus. These changes were long-lasting beginning to return toward a normal profile only 5 hours after the end of shocking. Ribosomal dimers were not formed *in vitro* but existed *in vivo* in rats as proved by several methods. It was established by a variety of methods that the increase in ribosomal monomers and dimers did not result from the action of RNAse on polysomes. Status epilepticus ribosomal monomers and dimers did not bear polypeptide chains and thus were probably not actively making proteins and cerebral protein synthesis in status epilepticus was inhibited.

Multiple ECS induced a highly significant decrease of the incorporation of leucine into brain proteins, even when anoxemia was prevented by paralysis and ventilation with O_2.

Factors of mortality were investigated in freely convulsing cats and rats. Cardiac arrhythmias and circulatory shock , probably due to lactic acidosis, were the most common causes of death from multiple convulsions under the experimental conditions used. The role of lactic acidosis is particularly interesting since its correction led to a significant improvement in tolerance to multiple convulsions (preliminary study of 9 cats). If the animals were paralyzed and ventilated with O_2 during the seizure, 100% of ventilated rats survived a treatment sufficient to kill 100% of convulsing rats (30 consecutive

ECS), but when status epilepticus was maintained for long periods of time (1-3 H) there was a considerable mortality, even in paralyzed, O_2 ventilated animals.

The finding of long-lasting ribosomal changes in rats subjected to daily electroconvulsive shock and the knowledge that a small inhibition of protein synthesis is often sufficient to stop cell multiplication led the investigators to investigate the influence of seizures on brain development. Young rats received one ECS a day for 10 days either neonatally (days 2-11 of life), during the latter period of cell division in the brain (days 9-18), or after most cell divisions had ceased (days 19-28). Neonatal seizures significantly reduced brain weight and cell number. Slightly less immature rats exhibited a decrease in brain weight and cell size, but cell number was unaffected. In the largely postmitotic brain of older animals, neither cell number nor cell size were affected. These changes were not due to the electric current; the reduction in brain DNA was more severe than that due to malnutrition (for a similar reduction in body weight).

A quantitative study of pencillin-induced seizures has been initiated in another laboratory in order to explain the discrepancy between the pattern of seizure activity at the level of the hoppocampus by electrical stimulation or increased K+ concentration in CSF and the pattern of seizure activity induced by topical application to the hippocampus of crystalline penicillin G. High concentrations of penicillin and crystalline penicillin generated seizures characterized as a "spike afterdischarge." Subsequent studies with microelectrodes in similar experimental situations led to the discovery of the characteristic somatic discharge described as "paroxysmal depolarization shift." However, the studies, which also employed other methods for generating seizures (increased K+ in CSF, repetitive electrical stimulation), indicated that the leading role in the epileptogenic buildup took place at the level of the apical dendrites of the neurons, the soma becoming involved only in the immediate pre-seizure period. During these experiments and using chronic awake animals in which implanted cannulae permitted infusion of solutions over the dorsal hippocampus, the action of different concentrations of penicillin on hippocampal excitability was studied. The main result was that even in concentrations of 500 ugm/ml (i.e., 1,000 times less than the usual concentration used in most of the experimental studies) penicillin increased the epileptic excitability of the hippocampus. The buildup showed a pattern very similar to that induced by increased K+, with a gradual increase of the dendritic response, especially at the apical level. Spontaneous spikes (P.D.S.) appeared at concentrations five to ten times greater. As yet, sufficient data have not been obtained to determine the concentration at which the P.D.S. can be driven by afferent stimulation, or the concentration at which they become autonomous, discharging regularly with a relatively high frequency. The most important fact seems to be that different concentrations of penicillin can induce seizures in different ways by selectively activating some of the synapses of the involved neurons. The basic interest for a neurologist in search of a model representing human epilepsy lies in the study of epileptic foci where threshold is reached only occasionally when additional factors interfere. Future steps in this project will be directed towards analyzing the action and behavior of neurons in the transition between threshold epileptogenic concentrations of penicillin and supramaximal concentrations.

The penicillin effect has been studied further by another investigator. These discharges, resembling paroxysmal depolarization shifts, have been shown to respond to steady hyperpolarizing current injections in a manner which suggests that they are in part large EPSP's. Variable hyperpolarization, which constitutes the second phase of the response, is less clearly affected by hyperpolarizing current. Digestion by penicillinase removes the capacity of penicillin to produce paroxysmal discharges. The integrity of the beta lactam ring, therefore, appears to be critical for the excitatory action in leech ganglion as it is in cat cortex.

In another laboratory alumina, as a compound reacting with the brain, was investigated by histochemistry and electron microscopy to explore its convulsive properties. This showed alumina crystals fixed in monocytes, pericytes, fibroblasts and astrocytes near the injection site for years, but not within neurons as was formerly thought. Intracellular alumina in phagocytes is associated with increased fibrils and tubules and in many cells is associated with active appearing digestive phagosomes years later. Because exocytosis and disintegrating macrophages were not seen, it is suggested that small amounts of alumina may be broken down into soluble compounds which cause an intense fibroglial scar and convulsions. Neurofibrillary changes were not seen. The alterations seen thus far are ones of degree with the exception of axonal degeneration. Specifically, in small alumina lesions which do not produce seizures, minimal axonal degeneration is seen; however, these same small lesions do produce limited astrocytic changes with few neurons involved in the glial scar.

In order to lay the groundwork for a more detailed submicroscopic analysis, techniques for assessment of the extracellular spaces in the epileptic foci were developed. Techniques were evolved whereby horseradish peroxidase was introduced into the intercellular spaces. Initial studies indicate that this marker is extensively incorporated into synaptic vesicles under conditions when neurons are physiologically active, but not when neuronal activity is depressed by deep anesthesia. Thus, vesicles appear, for the most part, to be formed by micropinocytosis. If this preliminary observation is confirmed, the ultrastructural technique is available for documenting the turnover of synaptic transmitters and may provide the opportunity for a major insight of the structural substrate of the functional hyperactivity which characterizes epileptic foci.

In monkeys with chronic, recurrent seizures induced by intracortical alumina injection, degeneration in the brain has been studied by another investigator. Three to six months after creation of the lesion and after spontaneous seizures have been developed, axonal degeneration has been found which projects into the white matter of the ipsilateral hemisphere and into the thalamic nuclei with some degeneration traversing the corpus callosum. Degeneration in the putamen, seen at earlier stages, is no longer present. In chronic epileptic monkeys (2-5 years) axon degeneration is confined to the ipsilateral hemisphere and is seen predominantly in cortical association fibers, the centrum white matter and secondary motor, but not secondary sensory cortex. The main cortical fiber degeneration was largely in the inner and outer stripes of Baillarger. In similar monkeys in whom the seizures had been completely suppressed by anticonvulsant medication, studies undertaken several

months after complete seizure control showed little axonal degeneration. This indicates that continuing neuronal death occurs which appears to be related to the frequency and severity of clinical seizures. Electron microscopic studies of the fate of aluminum hydroxide give the impression that it becomes conjugated with extracellular proteins which are phagocytized by macrophages. Aluminum hydroxide crystals are rarely found in the cytoplasm of astrocytes and none is seen in neurons, axons or dendrites. Marked astrocytic proliferation accompanies this process. The aluminum, once injected, is not removed from the brain and does not appear to be detrimental to macrophages which contain it.

Exploring the potential of the alumina-cream monkey model for large scale evaluation of antiepileptic drugs, the efficacy of DPH, phenobarbital, and primidone is being tested in a 3 x 3 Latin Square design. Each drug is being evaluated in 3 monkeys at any given time. At the end of the study, 9 monkeys had received each drug separately for 6 weeks with four weeks of no-drug after each treatment. Three different dosages (for 2 weeks each) will be administered within each 6 weeks of treatment. Correlations among seizure frequency, blood drug level, and behavioral toxicity are being determined for each dosage of each drug.

Pharmacokinetics of carbamazepine in monkeys is being attempted as a prerequisite to evaluating the efficacy of this drug in the alumina-cream monkey model. Three normal catheterized monkeys are utilized. A range of 3 IV-bolus dosages will be repeated 3 times (with a week intervening between each) for each of the monkeys. After each IV-bolus, a series of frequent blood samples would be drawn within a 24-hour period. An attempt will be made to determine the shape of the drug distribution and elimination curve; whether it is biexponential; dose dependent; linear with time or with dose or both; and the drug half-life in monkeys from the slower component of the curve.

A technique used in another laboratory in inducing experimental seizures is based on sensory stimulation, such as elicitation of audiogenic seizures in susceptible mice and rats and photic seizures in rabbits and baboons. Interest has recently been directed to baboons which show a high incidence of spontaneous photic seizures comparable to those observed in photicsensitive epileptic patients.

The Senegalese baboon, Papio papio, as a model of an epileptic syndrome, has been considered similar to man. A very high percentage of animals respond to intermittent light stimulation with EEG abnormalities and tonic and clonic motor movements culminating in a major seizure. Until this model was discovered, pharmacological research into the important area of anticonvulsant compounds was forced to utilize electroshock or chemical convulsant techniques in rodents. The laboratory results from such studies have proved unreliable in predicting clinical efficacy of these drugs. More recent use of strains of mice, rats and rabbits with audiogenic seizures has also been disappointing in the predictive value of results to the clinic. The primate model of focal epilepsy induced with alumina-cream lesions has been valuable especially for studies of the epileptic process. However, because the brain development of the baboon is very similar to that of man, and because no local pathology can be uncovered

to suggest a focal lesion, this model appears especially relevant to the family of inborn non-focal epilepsies in man, particularly those of the photomyoclonic type.

About 60 to 90 percent of Papio papio show varying degrees of seizure-like responses to photic stimulation. It has been shown that the photogenic seizure is blocked by phenobarbital, dilantin, trimethadione, diazepam, and is enhances by chlorpromazine. Because of these findings this photogenic model has been considered as a valid analog of human epilepsy. Using this model, studies are now being conducted on (1) EEG events during spontaneous or evoked seizures, (2) study of new pharmacologic agents, (3) study of drug metabolism in the baboon, (4) study of neurohumoral changes in cerebrospinal fluid related to seizures, and (5) study of neurophysiological correlates of focal lesions in photosensitive baboons. More specifically, dosage regimens for diazepam and related drugs in adult baboons will be studied. Seizure development in infant and adolescent animals as well as the relationship between seizures and sleep patterns are being investigated. Neurochemical studies of CSF-content of 5-HT, epinephrine, etc. will be made by obtaining CSF samples before and during photogenic seizures.

Another research group has shown that the photosensitive epilepsy of the Senegalese baboon, Papio papio, could be desensitized by techniques similar to those developed for the clinical therapeutic conditioning of human photosensitive epilepsy. In addition, longitudinal study revealed the development of other neurological abnormalities. It is postulated that the baboons suffer from a slowly progressive degenerative encephalopathy (perhaps genetically determined), of which the photosensitive epilepsy is the most readily apparent manifestation.

Three new cases of "reading epilepsy" have been studied and the results obtained lend support to the concept that reading epilepsy is basically a communication disorder, and not a simple type of reflex epilepsy. Two of these patients were identical twins who had seizures when writing even when blindfolded. They also had seizures when playing the piano. When seizures were evoked by reading the seizure phenomena was typical of reading epilepsy, i.e. interruption of the reading process and minor jaw jerks. However seizures induced while writing or playing the piano involved clinically jerking of the hand or "freezing" of the hand in motion.

A study of the epileptogenic potential for photosensitive subjects of newer techniques of audio-visual display in modern television programming has been completed. It indicated that the projection techniques themselves are not effective stimuli. This is probably related to slow rates of flicker, short duration of the flicker and low intensity of the light, plus the alerting action of the accompanying images. A suggestion is made with recommendations to television producers for the avoidance of highly noxious visual stimuli and to parents of photosensitive children regarding television viewing patterns for their children.

One patient with an unusual form of somato-sensory evoked epilepsy was studied, in whom tactile stimuli applied about the forehead were effective in producing seizures, including application of EEG electrodes. This usually

happened prior to the time the electrode placement was completed and the EEG recording started. Two proved to be pseudo-seizures, and the other was pronounced musical hallucination in an elderly deaf woman. This latter patient is being intensively studied now.

Reflex epilepsy patients can be divided into two types; the simple type where the stimulus presented is simple, the response is immediate, the dysrhythmia is generalized, and where unilateral presentations are often innocuous. The evoked dysrhythmia is fairly generalized. By contrast, in the complex types the very nature of the stimulus is difficult to define. Examples of this are seen in musicogenic epilepsy where it is the theme of the music, or a subtle stimulus occurring during swallowing in the patient with seizures related to eating. The response is not immediate. For example, in reading epilepsy it usually requires several minutes of reading before the seizure occurs. The dysrhythmia is focal or primarily focal in the complex types.

The various types of behavioral treatment employed in the treatment of reflex epilepsy include (a) stimulus alternation, for example, use of monocular presentations in photosensitive patients or the employment of differential light intensity technique, (b) threshold alteration as in the musicogenic epilepsy and the voice induced epilepsy, where advantage is taken of the postictal elevation of seizure threshold, (c) vigilance trials, in reading epilepsy and epilepsy associated with eating, and (d) avoidance conditioning was used in a child with self-induced seizures who was intellectually incapable of cooperating in other forms of conditioning.

The role of biogenic amines in audiogenic seizures in genetically susceptible or resistant mice is being investigated in another laboratory. The preliminary work has suggested that animals genetically susceptible to seizures possessed decreased levels of serotonin and increased serotonin turnover. To study the role of monoamines in the process of seizure production, several experiments have been proposed. Various drugs will be employed to test the hypothesis that depletion of monoamines is associated with increased seizure susceptibility and that increased monoamines are protective. Reserpine will be used to deplete monoamines, L-dopa to restore dopamine, 5 hydroxy-tryptophan to restore serotonin, clonidine to stimulate norepinephrine receptors, apomorphine to stimulate dopamine receptors, and haloperidol to block dopamine receptors. Following the administration of these drugs or combinations thereof, assessment of seizure response will be made by placing animals in a glass dessicator fitted with a board which is mounted with an electric bell. The pattern of the seizure will be assessed in terms of "wild" running, clonic seizure, and tonic extension; assessment will be made both by the pattern elicited and the incidence of death. These drugs will be employed not only in the DBA strain at the susceptible age but as well in the primed-seizure in the C57 strain. Following the completion of these behavioral assessments biochemical changes in the brains of these animals will be measured in response to pharmacologic manipulation of the monoamines.

Clinical solutions to the problem of epilepsy are concerned primarily with medical and surgical approaches. Before surgery is performed, it is necessary to ascertain that the seizures originate wholly or in part from an area of the temporal lobes that can be safely removed without causing serious

neurological damage, the seizures occur frequently and are incapacitating, and presently available drugs are ineffective in controlling them. Also, the surgery must be accomplished so as to avoid creating a secondary scar which may in turn cause recurrence of the seizures. Recently, it has been found barbiturate and thiopental used in surgery can pinpoint areas of the diseased brain tissue responsible for epileptic seizures. Locating this tissue precisely permits it to be removed surgically.

The overall aim of another research program is the delineation of surgically removable lesions in severe, drug-resistant focal epilepsy, especially the psychomotor type, employing chronically implanted, intracerebral electrodes after conventional means have proven to be inadequate. EEG recordings are made of both spontaneous and stimulated potentials from chronically implanted, intracerebral electrodes in patients with focal epilepsy. The long-range goal is the telemetering, recording and analysis of data from patients in the non-hospital environment; i.e., at home or at work. Data are telemetered from implanted patients, as well as scalp EEG's from patients wearing a simple cap containing the electrodes. Data are obtained from continuous recording of EEG in order to record the onset of spontaneous seizures and the threshold of electrical stimulation at the subcortical sites. It is estimated that presently available techniques permit localization of the focus in one of four such patients, while this method, based on EEG studies, localizes three out of four such foci. Based upon the information received, a decision is made whether or not an operation is indicated. The operation is a standardized anterior temporal lobectomy in which the anterior 6 cm of the temporal lobe is removed en bloc. Of 29 patients operated in the past, 25 are seizure free and three have maintained a reduction in seizure frequency.

The treatment of epilepsy has depended to a large extent on drugs. One of the earliest medications was a sedative called bromide. This was followed by another sedative, phenobarbital, which worked better, but caused drowsiness in some cases. About twenty years ago diphenlhydantoin (DPH) (Dilantin) was introduced in the treatment of epilepsy. It has been widely used and has proved to be a valuable anticonvulsant drug. However, it was soon recognized that ataxia sometimes occurred as a complication of therapy with this drug. The ataxia may develop rapidly over a period of a few days, or insidiously over weeks or even months. One unexplained feature about the ataxia is the fact that occasionally a patient will rapidly develop ataxia in spite of having taken the same dose of DPH for several years. It has been thought that ataxia is a benign symptom only requiring a reduction in dosage or occasionally withdrawing of the drug. It was, however, later conclusively shown that cats subjected to DPH medication had developed severe loss of Purkinje cells in the cerebellum and cystic gliosis of the cerebellar white matter. A number of cases of permanent damage to the cerebellum, apparently due to this drug, have also been reported.

While studying the mechanism of action of DPH, it has been postulated that its major primary effect is to decrease membrane permeability to calcium in every tissue in which it is known to have an effect. Recently, one investigator has determined that DPH inhibits stimulus-coupled (potassium) norepinephrine H_3 release from brain slices. It has also been shown that procaine does the same, and that both procaine and DPH reduce calcium-45 uptake in brain

slices. Tetrodotoxin has no effect on release of norepinephrine H_3 or on calcium-45 uptake. These factors are consistent with the view that the DPH effect on norepinephrine release is related to its anticonvulsant effect, does not result from reduction of sodium conductance, and is probably mediated through an effect on calcium by the same mechanism as local anesthetics. In other words, DPH may exert a primary action on calcium and indirectly suppress sodium conductance. This latter possibility has now been evaluated.

In these investigations, calcium-45 movement was studied. The following results were obtained. Both DPH and procaine were found to limit calcium-45 uptake in resting nerves, but tetrodotoxin did not do so. Both DPH and procaine were found to reduce calcium-45 release from resting nerves, as well. DPH also was found to prevent the increase in calcium conductance which occurred during electrical stimulation. Results from these experiments suggested that both DPH and procaine decreased membrane permeability to calcium in resting nerves, and that DPH completely eliminated the intracellular accumulation of calcium which ordinarily occurs during stimulation. The present working hypothesis is that DPH occupies a binding site in the cell membrane which functions as part of a carrier system for transmembrane calcium movement, thus reducing permeability of the cell membrane to calcium ions. It is believed that calcium ions have a regulating role in the movement of sodium across nerve membranes. It is conceivable that DPH, like local anesthetics, exerts a primary action on calcium and indirectly suppresses sodium conductance, an effect previously investigated. Future work will be devoted to refining experiments with the DPH effect on calcium movement in brain tissue by working with synaptosomal preparations from brain, and also to determine if DPH inhibits stimulus-coupled acetylcholine release from superior cervical ganglia.

Neuropharmacological studies in experimental epilepsy in another laboratory indicated that the decrease in epiletiform activity following the administration of DPH and phenobarbital is accompanied by a 6-fold increase in cerebellar (CB) Purkinje cell (P-cell) discharge. If, however, cerebellectomy is performed, the antiepileptic activity of both drugs decreased. Additional evidence is provided for a cerebellar action of DPH in mice with genetic cerebellar degeneration. In these "staggerer" mice, the threshold for maximal electroshock seizures was not significantly different from that of normal litter mates, but more than twice the dose of DPH was needed for antiepileptic protection against seizures which followed supramaximal stimuli. These data indicate that the cerebellum is indeed a necessary substrate for the antiepileptic action of DPH.

A study of the mechanism of antiepileptic action of diazepam (Valium) indicated that diazepam exerts its antiepileptic action through a mechanism similar to that of DPH: i.e. decreased seizure spread occurring concomitant with increased CB P-cell discharge rate. This increased P-cell discharge rate and decrease in epileptiform activity has a rapid onset and short duration and appears to account for the usefulness of the compound in acute seizure disorders in man.

A study has recently been completed which has identified a compound with extremely potent antiepileptic activity occurring at quite low blood levels and is not accompanied by increased rates of CB P-cell discharge. Lidocaine-

HCl exerts a greater depression of epileptiform discharge than does DPH, phenobarbital, or diazepam. This action is rapid, prolonged, and occurs without depression of respiration, blood pressure, or EKG. Lidocaine causes a 200-300 percent increase in the threshold for electrically-induced cortical after discharge and again, this correlates with extremely low blood levels of the drug. In addition, the antiepileptic action of DPH appears to be synergistic with that of lidocaine, presumably due to different sites of action of the two compounds. It thus appears that lidocaine may be an effective agent for the treatment of acute seizure disorders in man, possessing distinct advantages over diazepam, including a synergestic action with DPH.

A project has been conducted on the actions of epileptoid anesthetics in limbic and sensory systems in acute and chronic cats. One agent, enflurane (Ethrane) has been described as inducing anesthesia and analgesia subsequent to induction of electrographic seizure episodes. The episodes occur despite the fact that the animal is flacid and does not display behavioral manifestations of the electrographic activity. This dichotomy between electrographic and behavioral activity may provide an excellent model for subsequent studies on the basic mechanisms of hyperexcitable conditions.

A major effort in evaluation of anticonvulsant drugs by another group of investigators has been to study the action of a new anticonvulsant--sulthiame. The absence of status epilepticus on abrupt changeover from DPH to sulthiame allowed the utilization of a rapid changeover in a double blind study on an outpatient basis. The relative absence of toxicity of sulthiame, with the exception of hyperpnea, anorexia and paresthesias was noted. The use of serum drug levels allowing correlations of seizure frequency, toxicity, and drug dosage in an objective manner has been utilized in the outpatient study. The sulthiame drug study is nearing completion with a total of 69 patients accessioned. Thirty-seven patients were dropped prior to completion of the study for a variety of reasons, with increase in seizure frequency the most usual. Twelve patients have completed the entire study and 20 patients still remain on the study. The sulthiame drug study has been extended for the evaluation of carbamazepine. A pilot study lasting approximately nine months is planned to precede a double blind outpatient clinical evaluation of carbamazepine with DPH used as the control drug. As in the sulthiame study, patients will be selected who have partial seizures, are over 18 years of age and have four or more seizures per month. The double blind study will take approximately 16 months to complete.

Another team of investigators conducting a clinical trial on patients who have a record of intractable minor motor seizures and one of the four kinds of symptoms: (1) absence or staring spells (momentary loss of consciousness); (2) a kinetic or "Salaam" seizure (loss of muscle tone in trunk or neck; (3) myoclonic jerks (involuntary muscle contraction); (4) a combination of the three where no one system is consistently dominant. Using the anticonvulsant drug, clonazepam, a dramatic and long term improvement occurred in a large percentage of these patients. Clonazepam was significantly effective in controlling seizures in approximately three-fourths of the patients. Results varied from a decrease by 50 percent to complete control. Additional advantages of this drug include low incidence of side effects, rapid action when taken

orally, and sufficient anticonvulsant efficacy to allow withdrawal or reduction of other drugs. Mental function improved in all patients where previous drugs could be reduced or discontinued.

A study has recently been completed by another group in which 35 patients who were on nitrazepam have been switched over to clonazepam. The patients suffered from a variety of seizure disorders ranging from petit mal and myoclonic seizures to grand mal and psychomotor seizures. The results indicated that clonazepam is equally effective as nitrazepam (Mogadon). Further study is under way in which clonazepam will be tried in patients who are intractable to standard anticonvulsant drugs, not merely as a cross-over or comparison drug to nitrazepam.

Dipotassium chlorazepate (Tranzene) is a carboxylated salt of nordiazepam. Nordiazepam is the major metabolite of diazepam, and is as effective in preventing seizures in experimental animals as is diazepam. Because nordiazepam has a significantly longer serum half-life (22 to 24 hours) than diazepam (6 to 8 hours) it was predicted that treatment with nordiazepam should give higher serum concentrations and thus prove more effective than diazepam in the routine oral maintenance treatment of epilepsy. Unfortunately, nordiazepam is not available for oral use. However, dipotassium chlorazepate upon absorption is decarboxylated to nordiazepam.

Currently studies are continued on several standard anticonvulsant drugs such as dilantin, diazepam, methosunimide, mephenytoin, (Mesantoin) primidone, etc. by exploring the stability of the binding ratio in individual patients over time, serum protein concentration (as differences in the amount of protein in the serum would influence the binding ratio), and to study the binding ratios of other drugs, primarily phenobarbital and salicylate, to see if high or low binding is a general phenomenon within a given subject or is specific for an individual drug.

Two laboratories are studying the effects of the ketogenic diet which has long been known to be an effective therapy for intractable childhood epilepsy. However, its use has been limited by low palatability; between 85 and 90 percent of the total caloric intake has to be provided as dietary fat. Recently, medium chain triglycerides (a mixture of the triglycerides of octanoic and decanoic acids) have been found to induce ketosis more readily than do long chain (dietary) fats. A diet containing 60% of caloric needs as medium chain triglyceride (MCT) has been devised. The aim of the present study is to determine the long-term anticonvulsant effect and acceptability of the MCT diet in a group of children with refractory epilepsy. The effects of the MCT diet are compared with those of the standard 3:1 ketogenic diet. The degree of ketosis induced by each diet is assessed by quantitative determinations of acetoacetate and beta-hydroxybutyrate in plasma. A search is made for an animal model in which an anticonvulsant effect of MCT can be demonstrated, to elucidate the mechanism of action of the MCT diet.

In the therapeutic trials, 9 children were treated with the 60% MCT diet, 5 with the standard 3:1 ketogenic diet. Trials on both of these diets have been carried out in 4 children. A clinic has been set up, in which children on the diets are followed at monthly intervals for review of seizure frequency,

weight, neurologic status and determination of side effects of therapy. In the four children who have completed trials on both diets, two had better seizure control on the MCT diet, while in one the anticonvulsant effects of the 2 diets were approximately equal. One could not tolerate the standard ketogenic diet long enough to determine its effect. Three children preferred the MCT diet, while one preferred the standard ketogenic diet.

The work of another investigator consists of two related parts. The first concerns evaluation of the usefulness of the "kindling" preparation for assaying antiepileptic drugs. Kindling involves the gradual recruitment of clinical seizures by repeated electrical stimulation of various cerebral structures in animals. This preparation is now being used for testing anticonvulsant agents, including the establishment of ED50 and TD50 levels and of dose-response curves. The second set of experiments is intended to evaluate the anticonvulsant activity of several cannabinoid substances in three types of preparations: the "Utah battery" of tests in rats, the kindling preparation in rats, cats, and monkeys, and the photogenic seizure syndrome in Papio papio. In a sense the maximal electroshock seizure test and the Metrazol seizure threshold test of the Utah battery will be used as validation procedures for the other two types of preparations. Cannabis is often cited as a potential antiepileptic agent. Unfortunately the antiepileptic properties of this drug have not yet been examined in detail under controlled circumstances. However, evidence has been presented which bears directly and indirectly on the question of the antiepileptic properties of cannabis. Some patients have reported some beneficial effects of their private use of cannabis in terms of amelioration of their seizure conditions. The rationale of these experiments is to examine the effects of cannabinoid drugs upon the clinical and electrographic manifestations of epileptic activity in the same animal and at the same time in order to obtain further insight into their possible antiepileptic action.

One laboratory is carrying out a follow-up study upon 148 cases of childhood epilepsy in which anticonvulsant therapy has been suspended for 5-12 years to determine the frequency of relapse and to discern any prognostic criteria. Seizures recurred in 36 (24%). There was no relation of recurrences to sex, race, heredity, puberty or seizure frequency. With an early age at onset of epilepsy and prompt seizure control the recurrence rate was 13 percent. It was at least twice as high in cases with a late onset, prolonged duration, and neurologic, psychologic or electroencephalographic abnormalities. Relapse rates were lowest in grand mal attacks (8%) and febrile seizures (12%). In psychomotor attacks the recurrence rate was 25%. The highest rate was in children with Jacksonian seizures (53%) and multiple seizure types (40%).

This study attracted much favorable attention on account of the long duration of the follow-up and the attention to detail as to seizure types, EEG results, and psychologic considerations.

The objectives of another program are the determination of detailed molecular structures of anticonvulsant drugs with clinical or laboratory-demonstrated potency against seizures typical of grand mal epilepsy, and the discovery of stereochemical principles responsible for their efficacies. Excellent progress has been achieved during the past years, with the complete

structural determination of the drug sulthiame and the as yet not-completely-refined structural elucidation of the anticonvulsant carbamazepine. Similar studies will soon be initiated on the anticonvulsant ethylphenacemide.

Previous structural researches have conclusively demonstrated that DPH and diazepam, two of the most important clinical anticonvulsants, and procyclidine and trihexyphenidyl, drugs with demonstrable laboratory efficacy against induced seizures of the grand mal type, share certain stereochemical features in their three-dimensional conformational molecular structures. All of these agents have two bulky hydrophobic groups which are situated roughly at right angles to each other and in each case occupy the same areas in space relative to the rest of the molecule, and all have an electron-donor situated between the two hydrophobic moieties. In addition, if the four molecules are superimposed so that the hydrophobic groups of each best superimpose on the equivalent groups of the others, a second electron-donating functional group in each occupies a similar position in space. These results have contributed weighty evidence that (1) the stereochemical characteristics which were postulated to be responsible for the anticonvulsant action of the drugs studied are in fact the necessary features, and (2) DPH diazepam, procyclidine, and trihexyphenidyl likely share the same mechanism of anticonvulsant action and act through the same receptor site.

The elucidation of the molecular structure of sulthiame has revealed that this drug does not share the stereochemical features of the others investigated. Therefore, it may be concluded that sulthiame does not have the same mechanism (nor, probably, site) of action as the above anticonvulsants. This finding lends independent support to the recent postulation that sulthiame performs its biological activity as an inhibitor of brain carbonic anhydrase. The other drugs are not inhibitors of this enzyme and work through quite a different (though poorly understood) mechanism. Concomitant clinical evaluation of sulthiame has revealed it to be an unsatisfactory drug. Thus, it appears that compounds with the molecular stereochemical features that have been demonstrated to be common to the other anticonvulsants work through a mechanism which is more efficient than that of sulthiame.

PARKINSON'S DISEASE

Parkinson's disease is a progressive neurological disorder of unknown cause affecting certain brain areas responsible for the control and regulation of movement. Estimates of prevalence range from one case per 1,000 to one per 200. Primarily a disorder of middle age or later, its prevalence is thought to be increasing with the increase in the average life span.

The onset of parkinsonism is ultimately characterized by tremor, rigidity, and bent posture. Intelligence is usually unaffected. In the fully developed disorder the face becomes the characteristic "mask" with a hard stare coming from unblinking eyes. The patient often sits motionless, rarely crossing his legs or folding his arms.

Standard treatment in the past consisted of physical therapy, a variety of drugs, and surgery. The treatments of choice are now thalamic surgery or a highly effective form of replacement therapy making use of large oral doses of L-Dopa. Although L-Dopa induces remarkable improvement in most parkinsonism patients, it has very unpleasant side effects and some patients do not respond. Therefore, a still better treatment must be found and the greatest long range contribution of the research on L-Dopa may be that, as the first direct lead to the real cause of parkinsonism, it provides an approach to the development of a much more effective form of therapy. Despite many recent developments, evidence indicates that the newer methods of therapy have not substantially increased life expectancy.

L-Dopa acts as a therapeutic agent in Parkinson's disease presumably by increasing the concentration of dopamine in the basal ganglia. L-Dopa therapy also increases dopamine levels in the hypothalamus and median eminence which may alter the secretion of releasing factors which control pituitary hormonal secretion. Investigations of the effect of L-Dopa on hypothalamic-pituitary hormonal control may be important in the early detection of adverse side effects of long term L-Dopa therapy.

A group of researchers has found that of fifty-one parkinsonian patients who have been on long-term L-Dopa therapy, six developed "start hesitation," a difficulty in initiating walking, as a side effect. This symptom can also occur in untreated Parkinson's disease. In some untreated parkinsonian patients, rigidity is present and the gait is usually slow with short shuffling steps, whereas in some L-Dopa-treated patients who develop start hesitation, the muscle tone is either normal or decreased and the gait is normal or minimally impaired once the patient starts walking. These investigations showed that patients with start hesitation had a significantly better response to L-Dopa therapy than the remainder of the L-Dopa-treated patients.

These investigators are also studying the possible relationship of catecholaminergic-cholinergic imbalance to the production of drug-induced extrapyramidal side effects in psychiatric patients by using the physostigmine test. The response to physostigmine is assessed by careful observation of the vital signs, neurologic status and handwriting. After a baseline physostigmine test, the patients are treated with phenothiazine tranquilizers as indicated by their clinical status. Results to date indicate that physostigmine has no

neurological effects on untreated patients and did not induce or aggravate dystonia, dyskinesia or akathesia. So far, they confirm the relationship between central cholinergic stimulation and tremor and rigidity. Dystonic reactions or dyskinesias after physostigmine injections have not been observed.

An abnormality in dopamine metabolism has been well documented in Parkinson's disease and implicated in Huntington's chorea. Both diseases are extrapyramidal disorders resulting from anatomical and functional abnormalities of the basal ganglia, which contain the highest concentration of dopamine in the brain. In Parkinson's disease, the levels of dopamine in the brain are decreased. L-Dopa, the immediate precursor of dopamine, is the most effective therapeutic agent for Parkinson's disease, presumably due to an increase in brain dopamine. These investigators recently obtained an IND from the FDA and university approval to study the biochemical and clinical effects of gamma-hydroxybutyrate (GHB) in patients with extrapyramidal disorders. Much of the initial investigation has been designed to determine absorption of the drug, plasma disappearance rates, toxicity, optimal dose, etc. Initial indications are that chronic treatment with GHB appears to be safe and well tolerated by patients and the investigators are encouraged by preliminary experience to undertake a thorough well controlled evaluation of this promising drug. In addition, GHB will be given a clinical trial in other neurological disorders such as Dystonia Musculorum Deformans, Spasmotic Torticollis and Tardive Dyskinesias.

A double-blind study was just completed involving 23 parkinsonism patients to evaluate the efficacy of amantadine in addition to L-Dopa. Patients were given either placebo or amantadine for two weeks and the alternate drug for the next two weeks in a double-blind crossover evaluation. Subjective reports from the patients, neurological examinations with quantitative scoring of the signs of parkinsonism, and timing of various activities were analyzed during each patient's visit.

During the next five months, L-Dopa was administered in slowly increasing dosage until either side effects or an optimum benefit was obtained. At the sixth month of study (five months of L-Dopa) a repeat double-blind crossover of amantadine and placebo was carried out while L-Dopa was continued at optimum dosage. This crossover tested whether amantadine continued to be effective, or; if previously ineffective, was now effective in the presence of L-Dopa. The better "drug" was selected to be given along with L-Dopa for the following five months, at which time a final amantadine/placebo crossover was carried out to study the effectiveness of amantadine after one year. The data is currently undergoing analysis.

Treatment of psychotic patients with reserpine or with phenothiazines has induced pharmacological parkinsonism which plays a cardinal role in developing modern neuropharmacology and modern treatment of idiopathic parkinsonism. Weaker psychotropic agents, such as the butyrophenones have induced at least a parkinson-like tremor if not also additional symptoms of parkinsonism. Conversely, treatment of parkinsonism with anticholinergic agents, L-Dopa, or both has induced pharmacological psychoses.

It has been considered by one team of investigators mandatory to determine in a conservative manner whether a cholinergic agent can be effective against

L-Dopa-induced psychoses without jeopardizing significantly the control of parkinsonism. Studies have been initiated of cholinergic agents in L-Dopa-induced psychoses. Physostigmine has induced severe aggravation of parkinsonsim, but only in a minority of patients. The majority experienced either mild or no worsening of their symptoms. Its worsening effect, furthermore, was apparently absent among parkinsonism patients receiving L-Dopa. Therefore, physostigmine was injected in four patients showing mental aberrations from L-Dopa administered together with a peripheral metabolic inhibitor. In the first of these patients injection of 1.0 mg of physostigmine caused near-total disappearance of both the mental symptoms and the involuntary movements exhibited at the time.

One patient was highly susceptible to pharmacological psychosis when given cholinergic agents or L-Dopa in amounts sufficient to affect her parkinsonism. This patient had not developed mental aberrations while receiving oral apomorphine, although her parkinsonism was improved markedly. The effects of physostigmine on her parkinsonism were restricted to reemergence of mild tremor and salivation, without detectable diminution of rigidity and akinesia. On the basis of these results, this team of investigators has petitioned the FDA for an IND permitting the study of chronically administered oral physostigmine.

Among 31 participating patients the following investigations were performed by injecting either apomorphine or placebo: (1) The injections were given double-blind to 17 patients with or without oral L-Dopa. (2) In 14 patients receiving L-Dopa, injections were given during predictable episodes of tremor, hypokinesia or involuntary movements, in most of them double-blind. (3) In 10 patients apomorphine was injected 3-6 times a day for 2-43 days. The patients in groups 1 and 2 were scored by a system for assessing their parkinsonian symptoms or their dyskinesia prior to and at frequent intervals following the injections. In group 3, these scores were obtained periodically.

With or without L-Dopa, apomorphine was effective first against tremor and rigidity, if present, but it also decreased the periodic bradykinesia in those receiving L-Dopa. Side-effects were not additive to those of L-Dopa. Indeed, the "awakening effect," the involuntary movements and even the nausea induced by L-Dopa were antagonized by injections of apomorphine whereas the sedative effects and nausea of apomorphine were antagonized by L-Dopa.

Previous attempts to use apomorphine orally in the treatment of parkinsonism have failed. Since L-Dopa can induce tachyphylaxis (tolerance) only in peripheral tissues and not in the brain, these investigators believed that one might be able to induce therapeutic effects with oral apomorphine, (while eliminating the expected side-effects) by following the precedent of L-Dopa. They administered gradually increasing oral doses of apomorphine to 14 patients with parkinsonism, two of whom were receiving L-Dopa. The others represented several stages in the progression of the disease. All were given placebo capsules for several days prior to and after interruptions of apomorphine administration.

The two patients who previously received L-Dopa plus apomorphine appeared to require less L-Dopa for similar effect. Five of the other patients exhibited significant, steady improvement of tremor, or rigidity and of akinesia. The lowest doses at which improvement emerged ranged between 160 and 600 mg/d and the fact that no drug was given during the night did not influence the progression

of the improvement. Improvement increased steadily with dose. These experiments have already proven that one can totally circumvent all side-effects of a catecholamine given acutely, by administering it slowly and gradually, thus inducing sustained cerebral effects.

The Schwab ergograph for the rapid measurement of changes in the control of parkinsonism has been automated by this same team of investigators. As a result, sophisticated motor data can be collected and routed to computers by relatively inexperienced personnel.

Neuropsychologists working in conjunction with members of a clinical neurology group have continued studies of cognitive and perceptual functions in parkinsonism. Attention has been directed to the performance of parkinson patients compared to a non-parkinson population, the role of cerebral dominance in relation to lesions of the striatum using laterality of parkinson symptoms as an indicator, and the effects of both short and long range administration of L-Dopa on perceptural motor testing compared with the administration of other anti-parkinson drugs. These investigators have compared the performances of patients with Parkinson's disease with that of controls on tests of motor ability. The performance of patients with parkinsonism was significantly worse than with controls.

Patients with primarily right-sided symptoms (inferred left hemispheric dysfunction) had bimanual alterations in both the tapping and the tracking tasks. Patients with primarily left-sided symptoms (inferred right hemispheric dysfunction) performed significantly worse than the controls on the tapping test only with the hand contralateral to the major brain damage. However, in the tracking task, where a visuospatial factor was required, the group with left-sided symptoms was more impaired.

As a result of these findings, additional studies were conducted to determine the effect of L-Dopa therapy on the performance of parkinson patients on these tests. The results indicated that, along with the neurological improvements observed on a clinical basis and compiled from the neurological rating sheets, the parkinson patients on L-Dopa therapy improved on two motor performance tests when tested after three months and one year after initiation of the therapy. Analyses of the data in terms of right-left errors revealed that aspects of cerebral dominance play a role. A substantial improvement in visuomotor coordination in a patient population who had had an average of 10 months on L-Dopa therapy was found. These data support earlier findings that sensorimotor tasks improve after L-Dopa therapy. It was also found that memory performance of parkinson patients is significantly inferior prior to L-Dopa administration and that there is a slight improvement after one year of L-Dopa therapy.

Reports have appeared in the literature that lesions of the ventrolateral nucleus of the thalamus or the pulvinar relieve hypertonia in humans. Since a cerebello-thalamocortical circuit has been implicated in maintaining muscle spindle excitability through fusimotor efferents, experiments were performed to study the thalamic contribution of spindle responses. Resulting data show that lesions in VLN but not in P depress tonic fusimotor activity, which results in a depression of the responses to stretch of spindle primaries.

The acutely decerebellate cat has been used as an animal model of hypertonia known to result from heightened alpha motoneuron activity in association with diminished gamma motoneuron activity. This model was selected to determine whether the alpha motoneuron hyperactivity results from increased responses to specific stimuli or from a non-specific heightening of responses to any stimulus. The investigators concluded that the limb hypertonia in this animal model results from the heightened effects of labyrinthine and joint proprioceptive influences on alpha motoneurons. Thus, the "release mechanisms" are highly specific and stimulus-related.

Other investigators have demonstrated that intraperitoneally administered L-Dopa predominantly enhances the responses to click stimuli recorded from the caudate nuclei of awake, freely moving rats whereas reserpine decreases the responses. These effects are contrary to the anticipated results, since previous studies indicate that dopamine, the metabolic product of L-Dopa, functions as an inhibitory agent in the caudate nucleus. L-Dopa has effects similar to reserpine on the responses recorded from the globus pallidus and the substantia nigra, consisting of mixed enhancement and depression of the responses. Thus, there are marked differences between the responses of the various regions of the basal ganglia to administration of monoamine-enhancing and monoamine-depleting agents.

Several theories have been advanced to account for the effects of L-Dopa in alleviating certain abnormal movements in man. These for the most part, were concerned with biochemical mechanisms only, and virtually nothing is known about the synaptic effects of L-Dopa on central neurons involved in processing sensorimotor activities. Two types of experiments were performed on the neuraxially intact squirrel monkey to obtain information on the effect of L-Dopa on subcortical synaptic organizations. The effects of i.p. administered L-Dopa were studied on evoked activities in VL and in the motor cortex (MC) during low frequency (8/sec) stimulation of corona radiata immediately below the MC. Following administration of L-Dopa, all evoked responses in VL and MC, except the antidromic potentials in VL, were attenuated or abolished. A major inference here is that CR stimulation, following administration of L-Dopa, orthodromically activates midline thalamic nuclei as effectively as localization midline thalamic stimulation does.

The data show that a MC VL-midline thalamic inhibitory pathway is one of the subcortical target sites of some metabolites of L-Dopa. It has been reported that L-Dopa elicited 8-14 sec waves in the flat EEG of humans in coma following head injury. It was deemed desirable to study the effects of L-Dopa on apparently silent ECoGs. Temporary and repeated rotations of the trachea and mediastinum of the gas-anesthetized monkey results in the rapid development of flat ECoG. Observations on effects of L-Dopa in VL and MC were carried out following mediastinal rotation in acute experiments lasting up to 34 hours. The experiments revealed that dopamine or other metabolites of L-Dopa exert powerful effects on intrathalamic synaptic systems in the monkey.

MULTIPLE SCLEROSIS

Multiple sclerosis (MS) affects approximately a half million people in the United States and is considered to be one of the major neurological disorders. The onset of the disease occurs predominately in the 20-40 year old group and is usually progressive for the remainder of the life span, although intermittant remissions lasting months or years are commonly experienced. The symptoms are quite variable depending upon the part of the central nervous system affected, but the general pathological findings are fairly constant, involving demyelination of white matter and subsequent plaque formation. The etiology of the disorder is unknown although many investigators assume it to be either viral or immunological. This hypothesis is primarily based on information obtained from other demyelinating diseases. However, since myelin is a product of neuroglial cells, an aberration of glial metabolism is considered to be involved.

Still unexplained is the unusual geographic distribution of MS. The incidence increases significantly with latitude, the highest occurring in the north temperate zones of Europe and North America. Adult migrants to areas of relatively low incidence, such as Israel, retain the pattern of their native region, while children under 15 years of age assume the lower risks characteristic of natives of the country to which they have immigrated.

One investigative team has been exploring the possibilities that MS and ALS are the results of immunological responses. Although there is little evidence for this in ALS, there is some to indicate that an immunological mechanism is operative in MS. To date this group has tested 31 different sera for their effects on anterior horn cells in tissue culture. All of the ALS sera destroyed cultured anterior horn cells and none of the sera derived from other pathological sources had any obvious effect on these cells. It is notable that MS sera did not destroy anterior horn cells. This work is continuing.

One of the central questions in research on demyelination concerns the mechanism of myelin breakdown. Currently this avenue is being explored by this team using both quantitative histochemical techniques and fluorometric methods.

This same group is the first to institute the use of large scale preparations of normal oligodendroglial cells isolated from fetal and mature CNS white-matter for both tissue culture and metabolic studies. They have refined their tissue culture procedure to give an accurate, reproducible, predetermined number of oligodendroglial cells attached to the plastic well bottoms of Microstatic Tissue Culture Plates. These containers proved to be satisfactory for establishing an adequately controlled experiment in a single culture flask.

One research center has played a major role in defining the neurochemical changes in genetic disorders of metabolism that affect the brain. These genetic disorders have primarily involved abnormalities in the metabolism of sphingolipidoses, such as Tay-Sachs disease, and of gangliosidases. Upon the identification of the specific enzymatic defect, it

becomes possible to identify the disorder in utero. An investigator at the center has recently identified the enzymatic defect in Krabbe's disease; other sphingolipid glycosidases are being investigated to determine their roles in human disease.

Another genetic chemical disorder of the nervous system, tricopoliodystrophy, was found to be sex-linked. This disorder is due to a lack of copper absorption in the gut. It was found that cytochrome oxidase was present at less than 10% of the normal level in all tissues examined. Further, the younger the baby, the higher the content of cytochrome oxidase, possibly because of transplacental transfer. Intravenous copper therapy, while unable to reverse the disease process, resulted in a dramatic increase in mitochondrial function in one patient with normalized cytochrome oxidase.

It has been demonstrated that there is a marked preponderance of one type (Type 1) of muscle fiber in several cases of benign congenital hypotonia, central core disease and nemaline myopathy. These findings suggest the possibility of a neuropathic etiology in these diseases which had previously been thought to represent primary disorders of muscle. From studies of genetic and inflammatory myopathies, the very existance of "primary" disease of muscle is now clearly subject to further scrutiny.

A tissue culture laboratory has been established where monolayer cultures and explants are maintained. Three strains of measles viruses from patients with subacute sclerosing panencephalitis have been isolated. A model for this disease has been developed in the hamster.

Extensive data on the activity of lysosomal hydrolases in leukocytes have been collected on one family each with G_{M1}-gangliosidosis type 2, G_{M1}-gangliosidosis type 0, and a-L-fucosidase deficiency and on 5 families with neuronal ceroid-lipofuscinosis (NCL). Activities of varicus peroxidases and of ortho-aminolevulinic acid synthetase and dehydratase in tissues and serum of patients with NCL and blood relatives have been measured. Serum levels of alpha-tocopherol in patients with NCL and their blood relatives have been determined.

The study of the possible role of the measles virus in some form as an etiologic agent in patients with multiple sclerosis is continuing. Titers of measles antibodies are being followed on serum and spinal fluid samples from patients with multiple sclerosis, Devic's syndrome, or acute MS, and a few patients with subacute sclerosing panencephalitis. The preliminary results indicate several patients with acute MS have elevated measles titers and elevated first zone colloidal gold curves. They have shown significant drops in titers and protein levels coincident with remissions or clinical improvement. Two patients with high first zone colloidal gold curves have shown reversion to near normal with clinical improvement. The data are still insufficient to draw any conclusions.

Blood samples were obtained from 56 patients with MS, 30 with optic neuritis (ON) and 100 controls, and determinations made of histoantibodies to measles. A statistically increased frequency of HL-A type 3 was found in MS patients as compared to controls. The implication is that the increased

measles antibody titers found by others in MS patients are related to the increased proportion of HL-A3 carriers.

Based on recent reports of establishment of chronic herpes simplex virus (HSV) infection in mice by means of intradermal (ID) injection in a footpad, groups of Swiss-Webster mice (age 5-7 weeks) were inoculated by this route with GD-7 virus, with DA virus, and with BHK-21-adapted DA virus. These animals are currently being observed and there have been no signs of disease after 6 months.

Another investigator has been studying the metabolism of demyelinating central nervous tissue in respect to myelin and oligodendroglial cells. She has found a new demyelination model; rats maintained with small amounts of triethyltin in their drinking water show extensive demyelination. Up to 50% of the myelin may be lost over a period of two months. Currently, she is comparing the three models of demyelination in an attempt to find common metabolic features which may be a common mechanism in all demyelinative diseases. In these three models the oligodendroglial cells are to be studied in respect to synthesis of myelin elements and mitochondrial function.

The ability of experimental allergic encephalitis (EAE) serum to produce a total inhibition of oligodendroglial differentiation and myelin formation has been shown to be complement dependent and heat labile. The nature of the myelin inhibitory (MI) factor resides in the 7s globulins and in the IgG fraction (DEAE separation). The time of action of the MI factor is under study. By introducing the factor at various periods after initiation of culture, it has been determined that inhibition will not occur after the tissue has been 6 days in vitro, i.e., at the beginning of myelination.

A parallel set of biochemical determinations utilizes S-35 labelled sulphate and its incorporation into cerebroside. This is totally inhibited by MI factor and rapidly begins once the factor is removed, i.e., disinhibition.

An extensive examination of sera from MS patients and controls is in progress. This will determine the incidence of the anti-glial and anti-neuronal factors in the various populations. An intensive study of a select group of MS patients, who characteristically experience relatively frequent exacerbations, will involve monthly sampling of serum over a period of years. This will demonstrate the relationship of the immunological factors to the clinical state of the patient.

In guinea pigs, intravenous treatment with antithymocyte sera (ATS) around the time of sensitization with basic protein in complete Freund's adjuvant with tubercle bacilli almost completely prevents EAE. Physical and histological signs of EAE appeared in control animals although disease in the normal rabbit serum treated guinea pigs was delayed one week (occurring around three weeks instead of two weeks). In no ATS-treated animals were neurological signs evident. An evaluation of serum treatment following sensitization, when physical signs are apparent, is currently in progress.

CEREBROVASCULAR DISORDERS

In the United States it has been estimated that approximately two million individuals suffer from cerebrovascular disease with an approximate death rate of 200,000 per year. It has been only within the last ten years that increased interest has developed in cerebrovascular disease, primarily through the stimulation provided by NINDS. In FY 1973 support for research in cerebrovascular disease was at the level of approximately $6,400,000, representing approximately 40 research grants and 26 clinical research centers, including 10 Stroke Acute Care Research Units.

Both hypoxemia (a reduction of the arterial pO2) and ischemia (a critical degree of low perfusion of the brain tissue) can deprive central nervous system cells of the oxygen supply which they require to function normally. Therefore, consciousness is lost when the arterial pO2 is reduced to about 30 mm Hg or when the cerebral blood flow (CBF) falls to about 50 percent of normal. These and other observations have led to the view that hypoxemia and ischemia share equally the potential of inducing brain anoxia and that irreversible anoxic damage may be precipitated by either. However, recent experimental results have indicated that there is a fundamental difference and that permanent cell damage is probably almost always caused by ischemia.

It has long been known that the mature brain uses glucose as its main or sole substrate. In some tissues of the body the cells can live on the energy which is set free when glucose is anaerobically degraded to lactic acid. Since the adenosine triphosphate (ATP) yield of glycolysis is only about one-twentieth of that obtained when glucose is oxidized to CO_2 and water, the brain is constantly dependent on a continuous supply of oxygen. Under normal conditions, the rate of energy production in brain is adjusted to the energy use rates of production and utilization of ATP which constitutes a delicately poised energy state.

The effects of brain ATP levels on electrocorticographic and pathologic changes in focal transient ischemia in the squirrel monkey have been studied by a group of investigators. Cerebral ATP and lactate concentrations were measured in the squirrel monkey before, during, and at selected times after various periods of middle cerebral artery occlusion. A gradual decrease in ATP to 55, 35, and 20 percent of normal and the increase in lactate to 7, 8, and 10 times normal after two, three, and four hours of occlusion, respectively, were reversible with restoration of blood flow. Electrocorticograms recorded in acute preparations indicated a potential for recovery. The correlation of lactate levels with previous determinations of blood flow in this preparation supports the theory that loss of autoregulation and "luxury perfusion" in cerebral ischemia result from localized metabolic acidosis due to accumulation of lactic acid.

Work in another center using the rat to study the effects of the anesthetic state on brain energy and acid base parameters found that the type and depth of anesthesia did not significantly influence the energy state of the tissue, as reflected by tissue concentration of phosphocreatine, ATP, ADP, or AMP. There was no indication that lactate concentration varied with the depth of anesthesia, and there were only small differences in the tissue lactate of

barbiturate versus non-barbiturate groups. In all barbiturate anesthetized groups, however, there was a lowering of lactate concentration with an increase in the intracellular pH. These experiments failed to indicate that volatile anesthetics inhibit electron transport or energy transfer reactions in the brain. Further experiments were performed to include effects on some of the parameters in the un-anesthetized state, in nitrous oxide anesthesia, and in phenobarbital, using light and deep anesthesia. The awake, unrestrained animals showed blood values of glucose of 5.2 mM/kg lactate, 0.09 mM/kg pyruvate, and an LP ratio of 6.1. The unanesthetized but paralyzed animals differed in that their pH was slightly lower at an identical PCO_2 and this appeared to represent a slight acidosis with reduction of bicarbonates from 27 in the normal to 22 in the normal paralyzed. Presumably an adrenalin reaction occurs in these animals with blood glucose values of 15.7 mM/kg, and elevation of blood lactate to 2.1 mM/kg, and an LP ratio of 15.

The question still exists as to whether energy-rich phosphate can be increased and provide possible protection to the brain in situations of stress such as might occur in the course of natural vascular disease or artificially during the course of a procedure such as carotid endartorectomy. Two groups of animals were studied, one maintained in a steady state of 32°, and the second group maintained at 25°. These were further subdivided into animals maintained at each of these temperatures for 30 minutes and again for 60 minutes. Respiration was controlled with PCO_2s maintained constant and blood pressure monitored for steady state maintenance throughout. In all groups ion energy phosphate stores and energy charge potential were demonstrated to be maintained at normal levels. Since the metabolic rate of the brain was decreased, as indicated by other data available in the hypothermic animals, it can be assumed that the mechanism for production of high energy phosphate was reduced in exact proportion to the decreased utilization of high energy phosphate with maintenance of normal stores of ATP and phosphocreatine.

In studies on anoxic anoxia, animals were maintained with 6 minutes of nitrogen breathing. At the end of this time they were switched to 100% oxygen, and now for the first time cardiac massage was produced to re-establish blood pressure till the animals established their own heart rate, and maintained their own blood pressures spontaneously. Groups were studied at 1 minute, at 2 minutes, at 3 minutes, and at 6 minutes during the hypoxic stress and following hypoxia, and were sacrificed at 30 minutes and 60 minutes. During the induction of hypoxia at 1 minute there was no change in blood glucose although brain glucose was decreased and lactate was increased. Phosphocreatine and ATP decreased, while there were corresponding increases in ADP and AMP. At 2 minutes the blood PCO_2 had further decreased to 17 with a bicarbonate of 15. The blood lactate was now elevated to 5, and LP ratio of 43 (normal 14) and brain lactate had now increased to 14 with an LP ratio of 160. Phosphocreatine decreased from 5.3 to 0.2, and ATP from 2.8 to 0.5. ADP increased from 0.3 to 0.7, and AMP increased from 0.02 to 1.1. Tissue glucose fell from 3.5 to 0.33. At 6 minutes ATP and ADP were undetectable in the tissue. Following recovery with cardiac massage, in 10 minutes phosphocreatine was essentially normal, ATP rose to 2.5 (normal approximately 2.8) while ADP and AMP were normal. At this same time the energy charge had completely returned to normal values.

Further studies on the primary determinants of cerebral viability following transient ischemic brain injury demonstrated that not only the efficiency of restoration of blood flow, but more immediately, the ability of the ischemic cell to extract available oxygen was imperative for the utilization in reparative purposes, i.e., the extent and reversibility of cellular derangement of energy metabolism. The investigators demonstrated that total cerebral ischemia in the anesthetized, normothermic dog produced rapid disappearance of cerebral ATP content with a half-time of 3.8 minutes. Complete restoration of cerebral flow was accompanied by recovery of cerebral ATP levels if those levels had not been allowed to fall below 20 percent of normal, which is considered to be the threshold of irreversible injury with respect to ATP generating mechanisms.

The relationship of extremes of blood pressure (hypo- and hypertension) to brain blood flow in stroke patients has been studied and the investigators demonstrated specific minimum levels of systolic blood pressure as being necessary for the maintenance of adequate CBF. Below the critical systolic level, focal neurologic symptoms occur and the EEG is altered. One group of investigators studying the EEG in cats following middle cerebral artery occlusion found that no substantial EEG changes occurred until five hours post occlusion when a progressive decline in regional oxygen availability began. By about the eleventh post occlusion hour the EEG in the same region had become flat, only after which did a substantial rise in intracranial pressure begin to occur, amounting to about 30 mm Hg above the level recorded at the time of occlusion. Terminally, there was a transient remarkable "luxury perfusion" of the ischemic region followed by a further rise in intracranial pressure to about 75 mm Hg at which time the EEG in the control hemisphere became flat, both pupils were fixed and dilated and the experiment was terminated.

Studies in another stroke center on the relationship of the EEG and CBF during carotid endarterectomy in the human demonstrated that a significant unilateral reduction of rhythmic 8-20 cycle/second activity in the intra-operative EEG usually does not develop during carotid clamping unless regional blood flow falls below 18 cc/100 gm/min. In such patients, the EEG changes can be corrected by the use of an internal shunt. Continuous EEG monitoring with intra-operative measurement of regional cerebral blood flow by xenon intracarotid injection technique was accomplished during 38 endarterectomies performed on 34 patients under general anesthesia. Ten had persistent focal EEG findings pre-operatively and throughout the operative procedure without additional changes during carotid clamping. The remaining 28 patients had no focal EEG findings pre-operatively or during the initial phases of anesthesia. These 28 were subdivided as follows:

1. Nine showed blood flow values greater than 30 cc/100 gm/min. during carotid clamping without EEG changes.

2. Nine showed blood flow values between 30 and 18 cc/100 gm/min. during carotid clamping (two of these had minor focal EEG changes which returned to normal, one spontaneously and the other after internal shunting) and

3. Ten showed blood flow values less than 18 cc/100 gm/min. during carotid clamping with all ten having focal changes on the side of clamping but records returned to normal within 2-4 minutes after internal shunting None had neurological deficit in the immediate postoperative period.

In cerebrovascular disease, in atherosclerosis of the brain arteries, and in cerebral thrombosis with positive clinical findings, there is a decrease in the speed of the circulation through the brain. In both senile and presenile dementia, the retarded circulation through the brain is striking. In these latter cases, the CBF is very much reduced and the mean value amounts to 521 ml., which is a reduction of 43 percent, as compared with that for the normal.

Autoregulation of regional cerebral blood flow (rCBF) to induced systemic arterial hypertension in 16 patients with varying neurological disorders was tested. Hypertension was induced by increasing the arterial blood pressure by an intravenous infusion of Aramine (50 mg in 250 ml of saline). Seven (Group I) of the patients had a mean increase in mean arterial pressure (MAP) of 32 mm Hg and had preserved autoregulation while 9 (Group II) with a 56 mm Hg increase in MAP showed some degree of loss of autoregulation. The response to 5% CO_2 inhalation was also tested in 14 of the patients (during CO_2 inhalation the MAP increased a mean 10 mm Hg in Group I and 25 mm Hg in Group II). Group I had a higher control or baseline mean CBF than did the group with loss of autoregulation (41 versus 36 ml/100 gm/minute, respectively). The group with loss of autoregulation also generally had more severe vascular involvement as demonstrated by cerebral angiography. The mean CBF response to hypercapnia was greater in the group with preserved autoregulation (51% versus 36% increase).

During Aramine infusion the MAP was increased by 38% in Group I and 59% in Group II. The mean CBF was essentially unchanged in Group I but increased 24% in Group II. Whether or not autoregulation is preserved could be related to: (1) the greater induced increase in MAP in Group II than Group I, (2) greater angiographic involvement with a lower baseline in CBF in Group II than Group I or (3) a direct or indirect influence of various cardiovascular factors. These studies encompass goals (1) and (2) above. In addition the effects of vasodilators on CBF were studied, with the hope that the patients' condition would be improved by drug therapy. Betahistine hydrochloride was evaluated in 6 patients. All patients except one had occlusive cerebrovascular disease. The mean control CBF of the group was 39.1 ml/100 gm/minute. The reactivity of the patient's cerebral vasculature was tested by determining the CBF response to 5% CO_2 inhalation. During hypercapnia the mean CBF increased to 57.8 ml/100 gm/minute. Fifteen minutes after the completion of the rCBF measurement during CO_2 inhalation, the patient was given 16 mg of betahistine orally. Forty-five minutes after the drug administration a third rCBF measurement was performed. The mean CBF for the group after the administration of betahistine was 38.7 ml/100 gm/minute. Because no change in mean CBF could be demonstrated 45 minutes after the oral ingestion of betahistine, 2 patients were evaluated 30 minutes and then again 60 minutes after the drug administration. In both patients the mean CBF did not change significantly 30 minutes and 60 minutes after betahistine. In summary these 6 patients failed to show a cerebral circulatory response to

the oral administration of 16 mg of betahistine. The cerebral metabolic rate for oxygen ($CMRO_2$) did not change significantly from the control value (2.80 ml O_2/100 gm/minute) after betahistine (3.09 ml O_2/100 gm/minute).

Additional work on CBF in another laboratory on the effects of sympathetic nervous system stimulation on regional CBF (rCBF) has provided evidence that this stimulation causes a decrease in rCBF, even though the decrease was minimal when compared to the decreases in blood flow in other regions of the head, such as the scalp, salivary glands, skin, bone and teeth. These same investigators have also shown that in cats with occlusion of one middle cerebral artery there is a transient decrease in catecholamines. They also studied the variations in water content of the brain following one-sided middle cerebral artery occlusion. The water content of the brain was increased in both hemispheres two to three days following occlusion. Brain sodium levels were also increased, beginning as early as four hours following occlusion. Sodium uptake remained elevated as long as three weeks after occlusion.

Work in another center on brain fluid volumes shifts in some stroke patients suggested that: (1) the majority of subdural effusions are initially bilateral and freely communicating; (2) in some instances serial studies have documented a certain sequence involved in the resolution of the effusion; namely, the bilateral communicating effusions decrease in size and no longer communicate freely. Resolution of the effusion appears to begin laterally and migrate medially with the parasagittal portion of the effusion persisting longer.

Data have also been collected on the chemical composition of subdural effusions at varying intervals during the course of the illness. As anticipated, there was a steady reduction in the subdural protein content.

The first "irreversible" change in stroke patients to be identified is, in one sense, outside the brain altogether. Respiration stops. Unless the subject is artificially respired the brain will not recover when CBF is finally reinstituted because the brain is being recirculated with unoxygenated blood. Therefore, attempts to identify early any respiratory deficients, and correct them, may enhance the patients' chances of recovery.

Impedance pneumography studies of respiratory rates and patterns in acutely ill stroke patients demonstrated that respiratory pattern abnormalities occurred more frequently during sleep or in the presence of depressed sensorium and in patients with severe neurological deficit. It appears that Cheyne-stokes respirations (CSR) is not always related to the presence of bilateral cerebral lesions and is not closely related to immediate prognosis. The presence of central neurogenic hyperventilation (CNH) indicates brain stem dysfunction and in patients with cerebral lesions is associated with transstentorial herniation and death.

The direct relationship of catecholamines to the cerebral vasculature has been investigated in rabbits. Here, the responses of rabbit subarachnoid arterial smooth muscle, as contrasted with arterial smooth muscle from kidney, mesentery, lung, heart and skin, were studied. Smooth muscle strips from rabbit subarachnoid arteries did not respond to catecholamines in concentrations which caused strips from skin, mesentery and kidney vessels to contract. The strips from subarachnoid arteries responded to serotonin, histamine and angiotensin II but not to bradykinin, acetylcholine, methacholine, and adenosine phosphate compounds. Heart and lung vascular strips did not respond to catecholamines but responded to acetylcholine; cardiac strips responded to dilute methacholine. Lung strips often failed to respond to histamine in usual concentrations but did respond to bradykinin. The subarachnoid noid strips had a distinctive pattern of response as compared to vascular tissue from other organs. These observations indicate that, in vitro, cerebral vessels respond differently from vessels elsewhere in the body.

Further studies in another stroke center on 1) patients with unilateral hemisphere infarction in the area of the carotid artery and 2) patients with brain stem infarction, demonstrated in the latter group an increased ventilatory response to CO_2 which was significantly higher when a viscous resistance was used than during unloaded respiration. Both the ventilatory and inspiratory power responses were lower in these patients than in the others.

In the patients with unilateral cerebral arterial occlusion, the response of inspiratory power to CO_2 was .68+ .49 Kgmm before viscous loading and .77+ .55 Kgmm after. Both values were significantly higher than those obtained in the control subjects. Unlike the normal subject, the inspiratory power responses to carbon dioxide did not change significantly when the viscous airway resistance was added.

Even though many physicians and scientists would like to incriminate thrombi forming on top of or adjacent to atherosclerotic plaques as the major mechanism for the development of certain episodic cerebral vascular insufficiency syndromes, it is apparent that, in a number of instances, thrombi may not be demonstrated by pathological studies.

Until recently it was suspected that the fibrinolytic system was relatively inert, only becoming active under very profound stresses. The role of blood hypercoagulability in both the genesis of, and with respect to final outcome of, cerebral thrombosis, while extensively investigated previously, still is a controversial subject.

Pilot studies with plasma fibrinogen chromatography in the patient suffering from acute cerebral thrombosis demonstrated not only that blood hypercoagulability occurred commonly following acute cerebral thrombosis, but that both initial and serial plasma fibrinogen chromatographic data yielded important prognostic information. Essentially, if grossly abnormal blood hypercoagulable-thrombotic patterns were detected shortly after the acute ictis, and particularly if these patterns persisted, prognosis was poor. On the other hand, the finding of either a normal chromatogram or one indicative of enhanced plasma fibrinolytic activity was of good prognostic import.

Patients' bloods were classified into those showing normal plasma fibrinogen chromatographic findings and into those showing hypercoagulable/thrombotic patterns. "Normal" bloods, i.e., those with normal plasma fibrogen chromatographic findings, showed no significant changes in antithrombin III, alpha antitrypsin, alpha macroglobulin or plasminogen during the first five days of the hospitalization. In contrast, bloods with-hypercoagulable/thrombotic patterns showed significantly decreased concentrations of antithrombin III, alpha macroglobulin and plasminogen on admission to the hospital and for two to three days thereafter. The concentration of these moleties were at their lowest level at the time of hospital admission and therafter rose to normal levels within two or three days in the majority of patients. Concentration of alpha antitrypsin was approximately the same in both the normal and the hypercoagulable/thrombotic bloods.

These observations indicate that in patients showing hypercoagulable/thrombotic plasma fibrinogen chromatographic patterns, sufficient thrombin has been released into the circulation at the time of the ictus and subsequently, to significantly depress antithrombin III concentration. Similarly, the fall in plasminogen concentration and the corresponding fall of one of its inhibitors, alpha macroglobulin, demonstrates concomitant activation of the plasminogen-plasmin enzyme system, suggesting the presence of disseminated intravascular coagulation.

Additional studies on abnormalities of blood elements, viz. effects of lead on red cell structure and their contribution to the genesis of strokes have been investigated by another center. Preliminary experiments dealing with the effect of small concentrations of lead on cell deformability have indicated that lead at a cencentration of 10^{-5} M will quickly cause the red cell to become completely stiff. This in turn may affect flow through small vessels.

Other workers have investigated the effects of anticoagulant treatment of patients with transient cerebral ischemic attacks for the period 1955 through 1969. Seventy-nine patients with transient ischemic attacks who received long-term anticoagulant therapy and 119 patients with transient ischemic attacks who did not were studied. Nineteen per cent of the treated patients and 40 percent of the untreated patients had a cerebral infarct in the period of observation, averaging about 8 years. Five percent of the patients receiving anticoagulant had an intracranial hemorrhage compared to 4 percent of the untreated patients who had intracranial hemorrhage. There was no significant difference in survival between the treated and untreated patients at one, three, and five years of observation after the first transient ischemic attack. There was a significant difference in stroke occurrence favoring the treated patients at one, three, and five years of observation. However, the largest difference was in the first month after the onset of the first transient ischemic attack. Assuming that there was survival free of stroke for one month, there was no significant difference in treated and untreated patients at one year or at five years.

These same investigators also demonstrated that endarterectomized carotid arteries of cats are regularly occluded by fibrin-platelet thrombi within 15 to 30 minutes following restitution of flow after endarterectomy. The incidence

and rapidity of occlusion are not affected by prior administration of aspirin, or of Coumadin when the prothrombin time was in the "therapeutic range;" however, adequate heparinization (2-3 X normal coagulation time) for as little as 4 hours following restoration of flow after the endarterectomy, prevented occlusion not only during the period of heparinization but for a 3-week period of observation thereafter.

There has been a continuing interest over the years to find and evaluate readily applicable atraumatic techniques for the detection of occlusive vascular disease in the carotid and subclavian arteries. Two techniques now under study in some of the stroke centers are: 1) opacity pulse propagation measurement and 2) thermometry. Results of studies utilizing opacity pulse propagation correlate well with the presence or absence of occlusive vascular disease of the extra-cranial arteries and show promise as an atraumatic method for screening of patients. Studies to further define the minimal degree of vascular stenosis detectable by these techniques continue and indicate at present that stenotic lesions of approximately 30% are recognizable.

These studies also include evaluation of the effects of cardiac arrhythmia and hypertension, together with the clinical correlates of these physiologic aberrations. A major limitation of this technique, the time required and accuracy (± 8 msec) of manual measurements, has been overcome by the development of an electronic time interval measuring system incorporating on-line presentation of simultaneous individual measurements. The preliminary results are encouraging and suggest that utilization of the two techniques in combination is more useful than the use of either technique alone in the prediction of arterial pathology. Results also suggest that combination of these two techniques may be useful in the problem of detecting arterial occlusive disease when it occurs bilaterally.

Limited awareness of cerebrovascular disease co-existing with, or resulting from other systemic diseases is another reason why the prevalence of cerebrovascular disease in childhood has almost certainly been underestimated. Although the relationship of cyanotic congenital heart diseases to cerebrovascular complications is well known, relatively little attention has been focused on sickle cell anemia, leukemia, intracranial sepsis, particularly meningitis, and trauma as determinants of childhood cerebrovascular disease. In a preliminary survey, data from one center suggest that intracranial vascular accidents are, in fact, relatively common in association with these major disease categories and deserve far more attention to determine the role of cerebrovascular complication in the overall morbidity and mortality.

The development and evaluation of screening techniques utilizing symptoms of transient ischemia to predict stroke and eventual outcome have been pursued by a number of stroke centers. One of these centers has shown that there is no difference in survival between patients aged 40-59 with three or more and less than three symptoms. Patients aged 70 and over with more than 3 symptoms had lower survival rates but the number of patients were too small to make reliable estimates over the total period of observation. The survival rates for blacks and whites showed very little difference between two groups under the age of 55 but a distinct difference between the ages of 55 and 69 and over the age of 70. The survival rate for Negro females aged 55 to 69 was

depressed. The screening tool was most effective in the age group 55 to 69 and least effective in the age group 40 to 54.

These preliminary analyses suggest that the interview screening tool did identify individuals with an increased risk of death during the following three years and was relatively more effective in identifying groups with an inreased risk of cerebrovascular than other causes of death.

The influence of hereditary factors in cerebrovascular disease has been investigated by studying the families of 80 patients with a clinical diagnosis of CVA. The frequency of CVA in parents and siblings of these patients was compared with the frequency in the family of the patient's spouse. The frequency of recognized predisposing illnesses to CVA including hypertension, diabetes and heart disease was also studied. The patients and the spouses were excluded from the study population. Analysis of the data obtained on 160 parents and 384 sibs of the proband and on 140 parents and 336 sibs of the spouse revealed a frequency of CVA of 10.7% and 8.6% respectively. This difference was not statistically significant. However, when the sibs and parents were analyzed separately the difference between the sibs was significant, suggesting the possibility that a small added risk of CVA existed for certain close relatives of a CVA victim. Besides an inherited tendency to CVA, other factors were considered to account for the difference in frequency of CVA. Age, family size, and differential reporting of illness failed to account for the difference. However, both hypertension and heart disease occurred with greater frequency in the sibs of the patient. When patients with these predisposing illnesses were excluded and those with CVA alone were compared, it was found that relatives of the patient and the spouse had essentially the same frequency (3.1% and 3.2% respectively). Moreover, hypertension and heart disease were significantly more common in the relatives of the proband. The excess of CVA in the sibs of the proband could, therefore, have been due to an excess of predisposing illnesses such as hypertension and heart disease, and no independent inheritance of CVA was demonstrated. In the absence of certain predisposing illness, close relatives of CVA patients appeared to have no greater risk of CVA than genetically unrelated individuals.

TRAUMA (HEAD AND SPINAL CORD INJURY)

The plight of civilian patients in the United States who sustain injuries to the brain, and more especially to the spinal cord, resulting in permanent damage and/or paralysis, has been a dismal one. To the existing 125,000 individuals chronically paralyzed in this manner, between 5,000 and 10,000 spinal cord injured patients are added each year. Added to this are 20,000 head injury deaths with approximately 15,000 who sustain permanent disability. Most of these patients are young, being between the ages of 18 and 25, of which 84 percent are young men. Compounding the personal tragedy of a group that remains largely depressed and nonproductive is the recent estimate of the Armed Forces that caring for each spinal-cord injured patient from the time of his injury to his death costs approximately $900,000.

Recently interest has been rekindled in basic research concerning the morphologic, physiologic, biochemical and vascular events occuring in the spinal cord and brain within minutes and hours of an injury that produces transient or permanent disability. Until recently, though, efforts to reproduce experimentally the lesions observed in human material have been variable, due to the use of different experimental models and to a failure to quantitate the amount of trauma. The scanning of large sections with low-power microscopes has revealed an unexpectedly high incidence of ischemic changes, many of which would not be evident on routine autopsy. Therefore the culprit or end-result of trauma resembles very closely the end-result of stroke, anoxia of central nervous system tissue, probably through reduced blood flow.

Recordings of cerebral blood flow, systolic arterial pressure (SAP) and brain oxygen consumption ($CMRO_2$) in brain injured patients have been made to evaluate the patients' status, outcome and eventual treatment. These data have shown that in acute brain damaged patients, intracranial pressure (ICP) usually is increased, occasionally to the level of SAP, but frequently it is normal, and the relationship of ICP to survival is still in doubt except at very high levels. Even then, brain function may be normal at a cerebral perfusion pressure (CPP) less than 10 mm Hg if the brain is relatively spared by the process that causes the intracranial hypertension. CBF may be decreased, normal, or increased, and the level of CBF correlates poorly with ICP except in patients with intracranial mass lesions. $CMRO_2$ is almost always decreased, but the cause is uncertain. $CMRO_2$ may decrease as a manifestation of primary damage to the cerebral hemispheres and/or the brain stem, or it may be due to secondary damage from decreased CBF produced by other causes. Autoregulation may be intact or defective among brain damaged patients and among regions in the same patient. There is poor correlation between autoregulation and survival. Moribund patients may have intact autoregulation. Measurements of CBF before and after administration of hypertonic mannitol demonstrate that edema can reduce CBF independent of ICP. No inference can be made about the status of the cerebral circulation from ICP measurements nor does ICP correlate with neurological status. For these reasons continuous measurements of ICP are of limited value in the management of severely ill patients without information on the cerebral circulation and brain metabolism.

The effect of ischemic and hypoxic hypoxia on the function of isolated brain mitochondria has also been studied. The results of these experiments

demonstrate that rabbit brain mitochondria are capable of respiring adequately and, therefore, are not irreversibly damaged even after 30 minutes of total anoxia. Heavy uncoupling or inhibition was not observed until 40 minutes. Since functional recovery in the rabbit submitted to this insult does not occur after 5 minutes of brain anoxia, there is a poor correlation between the energy state of the tissue and brain function after compression ischemia.

Studies in another head injury center demonstrated that impaired blood flow was a significant factor in both the balloon compression and the gunshot models. In balloon compressed dogs blood flow, as seen through a cortical window, had virtually ceased as apnea developed. In the gun shot injury in monkeys it was noted that all animals died in which the flow fell below 30% three minutes post injury. Simple decompression of the balloon compressed brain resulted in prolonged survival of vegetative animals but the administration of hyperbaric oxygen resulted in a few animals that appeared normal. Hemodilution had a similar result. The best results were obtained with a combination of hemodilution and hyperbaric oxygen.

The drug dimethyl sulfoxide was studied because it stimulates respiration, it acts as a diuretic like mannitol and urea, and it belongs to the cold protection and to the radiation protection series. They demonstrated that it is superior to urea in management of the brain compressed monkeys. It was then used in the spinal cord model in dogs and was again superior to urea and also to corticoids. It was about equal to hyperbaric oxygen. There was no synergism between it and corticoids or between it and hyperbaric oxygen.

These various therapeutic agents were then evaluated in a model of pure ischemia, infarction of the middle cerebral artery of the squirrel monkey. Dimethyl sulfoxide and hyperbaric oxygen were both effective but were not synergistic. Hemodilution on the other hand produced poorer results.

Additional studies on the relationship of head injury to intracranial pressure (ICP) have shown that when ICP in monkeys is elevated by subarachnoid saline infusion, no change in gray or white matter CBF was recorded until the pressure reached 74 mm Hg. Elevations beyond this point produced similar decreases in mean and compartmental CBF. With balloon inflation, an early and progressive reduction in mean and compartmental CBF occurred. With intravenous water infusion to produce cerebral edema, CBF of the white matter showed progressive diminution from the point at which ICP began to rise while CBF of gray matter tended to remain constant.

As with the declining stroke patient, the onset of pulmonary function changes in the head injured patient are also omens of a declining status. Head injured patients have been studied and managed on completely controlled respiration under neuromuscular blockade. It is believed that these patients would have survived otherwise. The alveolar-arterial gradients were plotted during these intervals and demonstrated a marked rise several days after injury and subsequent reduction of the gradient. This rise of the A-a gradient is thought to be due to shunting, but the factors involved in this increase of physiological shunting are not known.

Pulmonary studies in head injured patients by another group have demon-

strated that the recorded pulmonary changes were due, to a large extent, to changes in cardiac output (CO). A decrease in CO, as seen during cephalic compression, resulted in a larger number of alveoli being unperfused, and a decrease in perfusion to non ventilated alveoli giving rise respectively to an increase in dead space/tidal volume ratio (VD/VT) and a decrease in pulmonary shunting (QS/QT). The opposite occurred during elevation in CO, observed during spinal cord compression. QS/QT followed the changes in CO.

Similar studies in another head injury center have suggested that head injured patients in the serious categories show a marked hypoxia and abnormal alveolar-arterial PO2 gradient. These findings are attributable to both impaired lung transfer function and to increased venous admixture. They demonstrate a physiological response to hypoxia and a low threshold and high sensitivity to CO2 as a respiratory stimulant.

Attempts to correlate cerebral blood flow against rises in the partial pressure of carbon dioxide in very ill patients indicated a uniform rise in cerebral blood flow in all cases with a rise in the PCO2. On the other hand, in these same patients a rise in the arterial PCO2 did not show a uniform rise in CMRO2. Therefore, paradoxical responses of cerebral O2 metabolism appeared in spite of the uniform increases in flow after hypercapnea.

INFECTIOUS DISEASES

Infectious diseases of the nervous system include many types of illness caused by, or communicated by parasites, such as bacteria, protozoa, fungi or viruses. Studies of infectious diseases are primarily concerned with epidemiology and etiology of causative agents, their mode of transmission, host relationships, and possible ways to control their propagation.

Methods most commonly employed for recognizing the presence of viral agents in cell cultures include observations for cytopathic effects, hemaglutination, hemadsorption, and interference, fluorescence and electron microscopies. Immuno-fluorescent techniques are being used in diagnosing and studying brain inflammation due to viruses. Other studies are concerned with experimental encephalitis, the epidemiology of Eastern equine encephalitis, the effects of parasites on the nervous system, testing vaccines for protection against arboviruses, and the possible role of viruses in acute neurological syndromes in children.

Criteria of the responses of the experimental animal to viral infection are established by careful monitoring of the physiologic behavior of the CNS following inoculation with tissue extracts and whole cells derived from brains of patients with chronic encephalopathies. The early events of the experimental disease in animals have been detected by electroencephalography because the EEG record becomes abnormal before any other signs of diseases appear. These techniques are being perfected for clinical application. However, at present they permit the selection of diseases animals for study during initial phases of the pathogenic process. Before drawing any definitive conclusions about the nature of the disease produced, especially about its relationship to the original human disease, it is well to remember that a CNS disease produced in animals may not be identical with the diseases of the patient from whom the tissue was obtained. A dramatic example of species difference in host response is the effect of simian virus B. In man it induces a disease that is severe, and usually fatal, whereas for monkeys this virus is innocuous.

Subacute sclerosing panencephalitis (SSPE) is a degenerative neurologic disease of children and young adults. It is characterized by progressive mental and motor deterioration, myoclonic jerks, and coma. The patients become severely emaciated and die from intercurrent infections. The diagnosis established during the incipient stages often shows a personality disorder or mental retardation and the EEG shows slowing and dysrhythmia. However, high amplitude, low frequency synchronous waves do not develop until the patient exhibits myoclonic jerking. Spinal fluid proteins and cell counts remain normal or increase slightly during the entire course of the disease. Transmission of encephalomyelitis from humans to animals and further from animal to animal, producing symptoms typical of SSPE in the animal, has provided an important new lead in isolating and understanding the causative agent in SSPE. During the last few years, evidence of a relationship between SSPE and measles virus has been established. With the help of electron microscopy, tubular structures have been seen in the brain with SSPE which resembled the nucleocepsides of measles virus. Further investigation showed a high or rising titer of measles antibody in serum, measles antibody in cereborspinal fluid, and measles viral antigen in the brain of SSPE patients. On the basis of cytopathology, filtration, and serology,

measles virus has been isolated from patients with SSPE. These patients had no history of clinical measles, but had received live measles virus vaccine. Some had rubeola once in their life time. Several characteristics common to both measles virus and SSPE have been found. They include antigenic properties, host-range, cytopathogenic effects, temperature sensitivity, thermal inactivation and interferon production. Although basic similarities have been established, still unanswered are the questions of how and why the virus persists during the long period after the patient has recovered from measles and before he develops SSPE. Is the virus different in some way from regular rubeola? Will measles vaccine protect against SSPE? Is the isolated paramyxovirus a neuro-adapted strain of measles virus or a distinct but antigenically related virus? Is a second "helper" or "co-virus" necessary to produce disease? If such etiologic agents exist, how do they interact? How does virus reach the brain and spread within the CNS? How is it maintained for years if indeed it is measles virus? Are the usually high serum measles antibody levels noted in most SSPE patients a response to viral antigen in the brain and how is the virus protected from antibody? Does interferon play a role in limiting the disease? Is it possible to reactivate a latent or defective virus in the CNS?

Six SSPE strains have been isolated and examined in sufficient detail. They were found to be more neurovirulent for newborn hamsters and less reactive with neutralizing antibody than classical wild or vaccine strains of measles virus. Data suggest that the isolate "SSPE (1)" is also poorly reactive with antibody. Consistent possession of increased neurotropism and decreased reactivity among the SSPE viruses examined suggested that they were variants of measles virus of a similar nature. This hypothesis was supported by finding among plaque isolates from both wild and vaccine strains of measles virus variants which resemble SSPE viruses in neurovirulence and reactivity with antibody. It seems likely that the SSPE strains emerge as variants from a measles virus population under selective pressures in the patient. Although these variants may simply represent the most persistent viral genome, it is possible that those properties which they were found to possess in common, or some associated viral characteristic, allow such viruses to produce SSPE. At present there are no known markers of wild strain or vaccine strain measles virus that allow determination of which of these strains might have given rise to a particular SSPE strain. Attempts to distinguish between wild or vaccine strains as a source of SSPE virus will be further complicated by SSPE isolates probably being highly selected variants of a virus population which originally infected the patient. The exact relationship of SSPE virus to measles virus may be defined only after detailed biochemical and genetic studies. Reports have appeared which describe some chemical and physical properties of measles virus RNA and of RNA and nucleocapsids from measles virus infected cells. The comparisons of virus specified RNA's in measles infected cells with those in SSPE virus infected cells have revealed thus far only quantitative differences among six distinct species of RNA. The search for minor qualitative differences between measles and SSPE virus specified RNA of each species has not been completed. Studies are also underway comparing measles and SSPE viruses in terms of virion RNA and peptides, as well as virus specified peptides of infected cells. Mutants of measles and SSPE virus have been isolated which are temperature sensitive and which have other markers that will serve in both biochemical and genetic investigations.

There exists today almost no cure for any of the numerous viral-induced neurologic diseases. In a few cases of CNS diseases, like SSPE, a virus has been isolated, although no therapeutic or preventive method has been conceived. It is necessary to know more about the physicochemical properties of the SSPE virus and its development in nervous tissues. Current efforts are directed to continuing the characterization of viral-specific nucleic acids and proteins from SSPE-infected cells, to attempting to detect the presence of virus in cultures through hybridization techniques or through identification of viral specific proteins and to continuing to isolate other degenerative and demyelinative viruses.

Although the etiology of multiple sclerosis is obscure, epidemiologic evidence has suggested a common exposure factor, most likely a virus, in those afflicted with this most prevalent human demyelinating disease. Studies of viral causes of demyelination have, in the past, taken two general directions: 1) in vivo and in vitro studies of animal viruses, and 2) attempts to isolate virus from human diseases such as multiple sclerosis, Schilder's disease and progressive multifocal leukoencephalopathy (PML), a disease in which papova virus-like particles had been observed by electron microscopy. The study of animal viruses causing demyelination has been limited by the paucity of experimental models. Work has centered on the three known viruses, distemper in dogs, JHM, a neurotropic strain of mouse hepatitis virus, and visna in sheep.

Canine distemper virus has been a difficult model to study because of the difficulty of obtaining and maintaining non-immune dogs. In infected canines, viral antigen has been found in both neurons and cells of the white matter. Perivascular demyelination follows the development of serum antibody suggesting the immune response may have a role in the pathogenesis of demyelination. However, demyelination in infected dog cerebellar explant cultures has supported a direct effect on myelin rather than an immunopathological process.

JHM virus induced demyelination in mice appears to be the most practical model to study. In 1949, Cheever and co-workers isolated a murine virus causing a disseminated encephalomyelitis with extensive destruction of myelin. The virus entitled JHM, also caused liver disease and subsequently has been grouped with the mouse hepatitis viruses (MHV) designated MHV-4. Mouse hepatitis viruses based on ultrastructure have been classified as corona viruses. A group of German workers produced demyelination in primates and confirmed the presence of the lesions in spinal cord, pons and cerebellum. This detailed histologic study shed little light on the virologic and immunologic aspects of the demyelinating process. Recently, demyelinative lesions have been observed in spinal cord of mice following neonatal thymectomy, but they were not identified as JHM lesions although electron microscopy clearly shows corona-like virus and the clinical course and histological lesions are identical to those induced in mice.

From the standpoint of human disease, attempts to transmit multiple sclerosis and Schilder's disease to experimental animals has, thus far, been unsuccessful. In vitro studies, although poorly documented, have also been futile. Progressive multifocal leukoencephalopathy (PML), the only human

demyelinating disease clearly associated with a virus has been of great interest. This subacute process, usually occurring in patients with imcompetence of their immune response, has been characterized by multifocal neurologic deficits. Patients have no fever or cerebrospinal fluid abnormalities and the course is progressive, ending in death. Multiple foci of demyelination surrounded by bizarre oligodendroglia have been found in the brain. Many of these cells have inclusion bodies containing papova virus-like particles. Attempts to isolate virus were unsuccessful until recently when a virus was recovered which appears unrelated to previously recognized papova virus.

Progressive multifocal leukoencephalopathy is caused by papova viruses, but their exact identity and significance in producing demyelination still require clarification. A strain has been isolated for which antibodies were detected in 60% of randomly collected human sera of all ages. This strain is distinct from polyoma and SV40 viruses. Tumors have been successfully induced in the brain of hamsters after intracerebral inoculation of this human papova virus. The research plan now includes: morphologic identification of the DNA in this and other papova viruses by means of the Kleinschmidt spreading technic; identification of viral antigens by immune electron microscopy: isolation and characterization of papova viruses derived from other cases of PML; morphologic studies on the formation of brain tumors induced in hamsters by human papova virus; and comparative studies of polyoma, SV40, and papova viruses from other laboratories.

Transmissible mink encephalopathy (TME) is akin to scrapie, kuru, and Creutzfeldt-Jakob disease, i.e., the noninflammatory, subacute spongiform viral encephaloppthies. These diseases develop over a period of months and years. The infectious agents are still unidentified, but their properties are distinct from conventional viruses. A study is being undertaken including sequential, morphologic study in hamsters and monkeys inoculated with TME brain suspensions in order to determine the localization and precise ultrastructural alterations that develop in neurons, particularly in synaptic regions, months before the onset of clinical symptoms. The proposed TME study is important in that the disease mimics human Creutzfeldt-Jakob disease. Thus far, all ultrastructural studies have dealt with the advanced stages of the disease in man an animal, but the elusive agent could conceivably be visualized earlier. The changes in the plasma membranes that appear to initiate the spongiform alterations in neurons warrant in depth study, for speculations concerning abnormal abnormal membranes are supported by biochemical data on the infectious agent.

Herpes simplex viruses, type 1 and 2, are the cause of life-threatening encephalitis in the United States today. The real frequency and clinical spectrum of disease is not fully known. Studies are limited by the current necessity of isolating herpes simplex virus (HSV) from the brain in order to make a definitive diagnosis. In preliminary studies passive hemagglutinating antibodies (PHA) in cerebrospinal fluid have been found early during the courses of several patients with HSV encephalitis and with Herpes virus hominis encephalitis. This PHA-CSF technique will be evaluated as an immediately available diagnostic test for HSV encephalitis. Cerebrospinal fluid PHA will be correlated with isolation of virus at brain biopsy and with measures of conventional and complement-requiring neutralizing antibodies in sera and CSF during various neurological conditions. Diagnostic studies will be augmented

by *in vivo* and *in vitro* studies of the pharmacology of potentially useful antiviral agents in experimental models and in man. An *in vitro* assay of antiviral activity in body fluids due to administered drug which is not affected by interferon or homologous serum antibody has been developed which includes concentrations of idoxuridine in serum, urine and cerebrospinal fluid of patients with suspected diagnoses of *Herpes virus hominis* encephalitis. Minimal inhibitory concentrations of agent vs strains of HSV will be correlated with amounts of drug in blood, spinal fluid, brain and urine after therapy. Possible antiviral synergy of several antiviral drugs in combination will be assayed. These data will guide rational therapy in man.

A number of viruses have been implicated in the etiology of multiple sclerosis. It is surprising that vaccinia infection, almost universal, has not been sought as a possible agent, but in fact, no studies on vaccinia studies of multiple sclerosis have been done, with the exception of electron microscope searches for elementary particles which have been unsuccessful. It is clear that if vaccinia antibodies are found in a significant number of spinal fluids of multiple sclerosis victims and in no other chronic neurologic disease, this might be of some diagnostic use for neurologists faced with early stages in multiple sclerosis, such as optic neuritis which may or may not progress to the full-grown picture of multiple sclerosis. More important, if vaccinia can be related to etiology, a significant reduction in incidence of routine vaccination may lead to a reduction in this disease. A study is being conducted on the frequency of the presence of neutralizing antibodies against vaccinia virus in the spinal fluids of victims of multiple sclerosis and of other patients suffering from chronic brain syndromes. Preliminary evidence suggests that only multiple sclerosis patients have such antibodies in spinal fluid. In order to relate the possible etiologic significance of this finding to the disease itself, it is proposed to do fluorescent antigen studies on brains of multiple sclerosis victims using specific antibodies against the following four soluble antigens of vaccinia (NP, S, HS, HL).

Visna is the only model of a slow, virus-induced demyelinating disease in animals, but work on this infection has been limited because the disease has only been produced in Icelandic sheep. Previous attempts to adapt visna virus to other breeds of sheep or other animals have not been successful. Because of the importance of this model to study demyelination relative to multiple sclerosis, further attempt have been made to adapt the visna virus. Preliminary studies have been carried out using the rapid serial passage of visna virus in fetal sheep. Intracerebral inoculations have been carried out by laporotomy at 50 days gestation in fetal lambs. Lambs were sacrificed at weekly intervals thereafter for virus titration on sheep testicle tissue cultures and for explants of brain tissue. Preliminary, but encouraging, results have been obtained indicating there is definite replication of visna virus in the brains of American sheep.

Visna was originally recognized in Iceland, as a naturally occurring fatal central nervous system disease of sheep, and was described by Sigurdsson as one of the prototypes of slow infection. The causal agent is now classified as an oncorna virus, based on its physical, biochemical, and biological properties. Following intracerebral inoculation os susceptible Icelandic sheep, a persistent infection develops followed by a late appearance

of neutralizing antibodies. Clinical disease appears in 6 months to years after inoculation, and is characterized by a severe lymphocytic infiltration of leptomeninges and choroid plexus, with marked perivascular cuffing and eventual destruction of white matter.

The study postulates that the disease is immunopathological in nature and will attempt to elucidate: (1) the cellular sites of virus replication; (2) the role of the immune response (antibody and cell-mediated) in disease production; and (3) the mechanisms of virus persistence. Approaches will include (a) a sequential study of infection in Icelandic sheep; (b) the effect of immunosuppression, using anti-lymphoid serum or cyclophosphamide; (c) the effect of active immunization before and after inoculation; (d) the correlation of various measures of antibody and cell-mediated immunity with disease progression; (e) a search for evidence of an auto-immune response against myelin antigens; and (f) the influence of age at inoculation upon the course of infection.

The main objectives of another program are to study the biological properties of slow neurotropic visures and their interaction with neural tissue at the cellular level, to further analyze the nucleic acids and proteins of visna virus, to determine if visna and maedi virus nucleic acids show complementarity to each other and to oncogenic RNA viruses, and to define the nature of the two types of extracellular particles observed in visna virus-infected cell cultures.

The results of the analysis of visna virus proteins suggest the presence of eleven to fourteen polypeptides in the visna virion, including at least two glycoproteins. Treatment of the virions with the proteolytic enzyme, bromelain, resulted in removal of most of the glycoproteins as well as some of the polypeptides apparently not containing carbohydrate. Two carbohydrate-containing proteins were completely removed by the enzyme treatment, suggesting that they were surface compounds of the visna virion. In the number and general pattern of their mobility in gels, the proteins of visna virus resemble the proteins of the RNA tumor viruses. These findings lend further support to a close relationship between visna virus and the RNA tumor viruses.

Examination of visna virus infected sheep choroid plexus cells after indirect ferritin labeling demonstrated that antibody bound to the surface of both larger extracellular particles lacking a central core and small particles containing osmophilic cores. In addition, crescent shaped buds emerging from the cell membrane were discretely labeled with ferritin. These findings indicate that the crescentic buds appearing at the surface of visna-infected cells as well as the larger spherical extracellular particles without osmophilic centers are antigenically related to visna virus.

Visna and maedi viruses appear to share common nucleic acid sequences, but equivalent annealing could not be demonstrated in two mammalian RNA tumor viruses. These results reinforce the belief that visna is closely related to maedi virus and may be a neutotropic variant of the viruses of the progressive pheumonia complex.

The results of these investigations may develop new approaches and techniques which will be of use in identifying cryptic viral agents as the causes of human subacute or chronic neurological diseases. They may also explain the pathogenesis of virus-associated demyelination and indicate similarities between the evolution of slow viral infections of the nervous system and the development of virus-induced tumors.

Muscular Dystrophy

Muscular dystrophy includes a group of diseases characterized by the progressive wasting of skeletal muscles. The condition is primarily genetic in origin, may be X-linked, affecting approximately five times as many males as females. The most prevalent disease entity encountered is that of childhood or the Duchenne type. Diagnosis is made from the medical history, microscopic examination of biopsy material and electronmyography. Patients and approximately 70% of carriers have elevated levels of serum creatine phosphokinase. The laboratory investigation of this disease is facilitated by the occurrence of muscular dystrophy in certain strains of chickens and mice.

The primary defect is believed to reside in the muscle itself although alterations have also been reported in nerve and at the neuromuscular junction. The "white, fast" muscle is primarily affected while the "red, slow" muscle is relatively resistant to dystrophy. Since these properties are, at least in part, determined by the nerve innervating the muscle, there is some reason to suggest that the peripheral nerves may be directly involved.

Recent research in this area has been predominately concerned with the biochemical characterization of dystrophic muscle. There is as yet no evidence to support the contention that the muscle membranes are "leaky" or more permeable to small molecules since the plasma concentrations of amino acids and carnitine are normal. The adenyl cyclase activity of dystropic muscle is normal, but the enzyme does not respond to epinephrine or sodium fluoride to the same extent as in normal muscle. Mitochondrial respiration appears to be unaffected while the ability of isolated sarcoplasmic reticulum to concentrate calcium is impaired. The capacity of isolated actomyosin to undergo "superprecipitation" is unchanged although actomyosin ATPase activity is significantly decreased.

Of potentially great importance is the observation that polyribosomes isolated from the muscle of patients with the Duchenne type of muscular dystrophy synthesize an excessive amount of collagen. Collagen synthesis is returned to normal if the polyribosomes are incubated in the post-ribosomal supernatant from control muscle. Yet to be uncovered are the substances or enzyme systems in the supernatant which control polyribosomal protein synthesis. This biochemical defect has been successfully used as an assay method to detect female carriers of X-linked Duchenne muscular dystrophy. The procedure uncovers all "true and probable" carriers (two or more sons with the disease, or other male relatives affected) and most "possible" carriers (only one male relative with the disease). Excess polyribosomal collagen formation is observed in preparations derived from approximately fifty percent of "possible" carriers.

This biochemical abnormality is also found in cultured skin fibroblasts suggesting that a simple, convenient and relatively painless biopsy procedure can be derived to detect carriers. It now appears possible that most or all carriers of Duchenne muscular dystrophy may be detected by a combination of

assays which include polyribosomal collagen synthesis, light and electron microscopy, and determination of serum creatine phosphokinase levels.

Myasthenia Gravis

Myasthenia is a chronic disease characterized by skeletal muscle weakness that may range in severity from slight depression of maximal strength to complete motor failure. These extremes of functional impairment may be seen at different times in the same patient and may range from slight ptosis to respiratory failure. The course of the disease is highly variable and the prognosis is uncertain. The mortality rate is estimated to be fifteen times that of the general population and the incidence approximately one in twenty to forty thousand. There is little evidence to suggest that the condition is genetically transmitted. The age of onset is usually 10 - 40.

There is little doubt that myasthenia results from a defect in transmission across the neuromuscular junction. It is not known for certain whether the condition results from the inadequate release of acetylcholine from the nerve terminals or from a decreased sensitivity of the motor endplate to a normally effective amount of the transmitter. It has been established that the size of the miniature endplate potentials, arising from the spontaneous continual release of acetylcholine from the nerve ending, is greatly reduced in myasthenia. On this basis some authorities believe that the condition primarily results from an inadequate release of transmitter from the nerve terminals. Definitive studies have yet to be done on the content, release, and resynthesis of acetylcholine in the nerve terminals innervating normal and pathological human muscle. Similarly, the sensitivity of the postsynaptic membrane has yet to be established. However, electron microscopic examination of biopsy material reveals changes in nerve fibers, muscle endplates and muscle fibers.

Evidence is accumulating that myasthenia gravis may be an autoimmune disease - the immune mechanism affecting the process of impulse transmission at the neuromuscular junction. This is the rationale for thymectomy, advocated by some for all patients. It is known that the IgG fraction of some myasthenia sera bind to the I-band of skeletal muscle which is composed mainly of a complex of troponin and tropomyosin.

Drug therapy in myasthenia is based either upon increasing the available acetylcholine at the neuromuscular junction or upon suppression of the autoimmune mechanism. In the former case, anticholinesterase agents are used which decrease the rate of destruction of the neurohumor at the endplate. The oral administration of relatively long acting, reversible inhibitors are employed which do not readily cross the blood-brain barrier. The systemic administration of short acting anticholinesterases are used for diagnostic purposes to ascertain whether a significant transient increase in muscle strength is observed. The efficacy of ACTH and corticsteriods in myasthenia is thought to be based on suppression of the immune process. Myasthenic patients are abnormally sensitive to a variety of drugs such as central nervous system depressants (morphine and barbiturates), tranquilizers, local anesthetics, quinine and some antibioties which effect release of acetylcholine. Of clinical importance, is the problem of over medication with anticholines-

terases, which also produces neuromuscular block.

Recent research in this area has largely centered about the development and testing of new pharmacological agents. As noted above, the use of corticosteroids is currently under investigation. It has been found to be especially effective after thymectomy.

Theophylline compounds such as aminophylline and oxtriphylline are under investigation. Their use is based on experimental evidence indicating that cyclic adenosine monophosphate is involved in the process of neuromuscular transmission. These drugs inhibit phosphodiesterase and thereby increase the level of the cyclic monophosphate.

Germine monoacetate, a semisynthetic derivative of the veratrum alkaloids is currently undergoing clinical evaluation. Its use is based on the production of repetitive electrical activity in motor nerve after a single normal action potential, resulting in increased neuromuscular transmission and muscle tension. However, the same mechanism apparently affects sensory nerves causing paresthesia as a side effect.

A potentially important development is the apparent production of an experimental animal with myasthenic symptoms such as muscular weakness. This was accomplished by the "hyperimmunization" of rabbits with homologous antigens. While neurophysiological and morphological studies has yet to be carried out, it is notable that these animals responded promptly to the administration of anticholinesterase drugs.

Hydrocephalus

Hydrocephalus is caused by mechanical obstruction of the ventricular outflow either within the ventricular system (noncommunicating hydrocephalus) or in the basal cisterns or at the pacchionian granulations (communicating hydrocephalus). In any case there is a disparity between cerebrospinal fluid production and resorption into the venous system. This usually leads to raised cerebrospinal fluid pressure and to progressive dilation of the ventricular system at the expense of the white matter of the cerebrum which becomes compressed. Hydrocephalus which develops in the adult is generally caused by mechanical obstruction in the circulation of the cerebrospinal fluid either by a tumor or by subarachnoid hemorrhage and subsequent scarring of the basilar cerebrospinal fluid pathways or by gliosis and obstruction of the aqueduct. Hydrocephalus is more common in infants and may also be caused by obstruction to fluid outflow. However, in the majority of cases no definite cause can be found and these infants with "communicating hydrocephalus" comprise the largest diagnostic group. The relationahip of cerebrospinal fluid pressure to dilation of the ventricals is not always clear since hydrocephalus in the absence of markedly elevated pressure is often encountered.

At present hydrocephalus is treated by various shunting procedures, most of which drain cerebrospinal fluid into another body cavity and thereby reduce ventricular volume. The many problems attendant upon the maintenancy of shunt patency in children are reflected in an estimated 25% success rate after fifteen years.

In animals, experimental hydrocephalus is produced by the injection of particulate matter such as kaolin into the cisterna magna. After one week cerebrospinal fluid pressure is greatly elevated and the animals display various neurological symptoms such as spastic quadriplegia, nystagmus and dysphagia. However, after three weeks cerebrospinal fluid pressure returns to normal and the neurological symptoms disappear. This is attributed to the development of alternative channels for cerebrospinal fluid absorption. It is postulated that the increase in cerebrospinal absorptive capacity results from transventricular fluid movement into the surrounding white matter and ultimately into the cerebral blood vessels. This is supported by the finding that the extracellular space and water content of the surrounding white matter is significantly increased.

The development of alternative pathways for cerebrospinal fluid absorption probably results, at least in part, from the increase in intracranial pressure. However in infants, the freely expansile nature of the skull often permits progressive ventricular enlargement in the presence of normal or slightly elevated cerebrospinal fluid pressure. This phenomenon can be duplicated in animals if large sections of the skull and dura are removed.

On the basis of these relationships a small number of selected hydrocephalic infants, one to three weeks of age, were treated by applying compressive head wrapping which prevented rapid skull enlargement and increased the intracranial pressure. This continued until the infants were six months old at which time the fibrous union of the cranial sutures limits the expansibility of the skull. At present these children appear to be developing normally and are free of the symptoms of progressive hydrocephalus, presumably as a result of the development of alternative pathways for cerebrospinal fluid absorption.

COMMUNICATIVE DISORDERS

When considering the several communicative disorders, our thinking is directed to the sensory functions and their attendent motor functions. There can be no expression without impression, no response without stimulation. A man does nothing, is not active, in any manner involving effectors for running, grasping or speech, unless in some way he is being influenced by energy changes occurring inside or outside of him which play upon his receptors - provided we except a few cases of smooth muscles and gland excitation by hormones.

Scientists interested in the phenomena of normal human nature and their prediction and control must have some definite knowledge as to how men are sensitive to influences, to what kinds of forces or influences they are sensitive, at what degrees of intensity, and at what places on or in the body the influences must be applied. No attempt systematically to understand the hows and whys of human behavior, such as communication, can be successful unless consideration be given to the paramount role of stimulation in the initiation and control of behavior. The National Institute of Neurological Diseases and Stroke has supported many researchers in sensory neurology and other aspects of sense modalities as it effects communication and its disorders. The current year's effort is replete with evidence of sturdy progression in the solution of many scientific problems which underpin the structure of our current knowledge of communication.

Neurosensory Instrumentation

A group of investigators has developed an instrument that allows precise, controlled warming of a local region of the skin from a radiant heat source. This would provide close regulation of temperature at the skin surface or heat energy transfer to the skin, and temporal and spatial characteristics of the stimulus. The approach is to use an infrared, carbon-dioxide laser energy source with a closed loop control via a radiometer detector which senses skin temperature.

This group is developing a remote microelectrode positioning and telemetry system for extracellular recordings from a single unit in the central nervous system of an unanesthetized, unrestrained animal. Another accomplishment was the development of a perfusion apparatus for continuously applying the proper fixative, under the proper conditions, to the human cochlea over the period of time required for dissection. Other investigators in this group are developing a technique for separation of neuronal and glial cells of brain tissue into highly enriched fractions and to separate out an enriched fraction of abnormal, atypical or diseased cells. In each case the fraction of undamaged cells must be sufficiently large for biochemical and metabolic studies to be made either directly or after a period in culture. Other investigators are refining a previously developed intercranial pressure monitor. This is an implantable sensor of intercranial pressure that requires no direct (percutaneous) extracranial connections and can remain implanted indefinitely. They further expect to develop associated extracranial equipment to determine pressure indicated by the transducer, to record, display and give warnings of dangerous pressure levels, and to be attached to the skull without harm or discomfort.

Another group of investigators reacted to widespread warnings of extreme risk to children's hearing when exposed to "loud" toys. Specifically, the consumer oriented magazines have on three occasions warned that the noise produced by the gunpowder caps of the WASP (X707) cap gun will deafen a child. While large peak sound pressure levels (155 dB) are indeed commonly measured, no relation to hearing loss has been reported. If a damage risk criteria is to be formulated for children's toys, it is necessary to provide some relationship between noise levels and hearing. Eleven adult listeners to one or five consecutive shots of the WASP cap gun fired by the listener with the gun held at arms length produced a measured temporary threshold shift at 4000Hz with recording attenuator and the Bekesy technique for 10 minutes following the noise.

Another laboratory is attempting to identify the cellular mechanisms involved in the etiology and pathogenesis of experimentally induced serous otitis media which leads to chronic inflammatory changes of the middle ear and mastoid. Middle ear effusion appears to represent a beginning phase in the continuum of inflammatory diseases of the middle ear and mastoid and the aim has been to study this precursor stage. The work began by establishing the normal ultrastructural, light microscopic and biochemical state of the middle ear of subjects (man, monkey, cat, and guinea pig) to which subsequent alterations were studied. It has also developed an animal prototype in which it could routinely induce experimental acute otitis media and the ensuing transitions to chronic middle ear effusion and finally to chronic granulomatous pathology and other chronic diseases of the middle ear and mastoid. While all this has been accomplished, it is currently engaged in corallary and parallel investigations of the mucoperiosteum and middle ear fluid of man and animals using biochemistry, light microscopy, electron microscopy, histochemistry and audioradiography. Acute experimental serous otitis media was induced in the squirrel monkey following Eustachian tubal obstruction. Oxidative and hydrolytic enzyme activity was generally increased throughout the mucoperiosteum. The periosteum showed a strong positive reaction for alkaline phosphatase. In this model, the passive transfer of serous fluid from subepithelial vessels to the middle ear was suggested by (1) chemical similarities of serous effusion and blood serum; (2) retention of carbon particles within the subepithelial small vessels in the presence of serous otitis media, and (3) ultrastructural evidence of basement membrane rupture and fluid flooding intercellular spaces, thereby distorting epithelial cells.

Another group of investigators are trying to determine if, in a patient with Meniere's disease (endolymphatic hydrops), an endolymphatic sac decompression and drainage operation done early in the course of the disease can conserve their hearing for several years (i.e., alter the rate of their hearing loss with time to that of the normal population). Ultimately, they will study 500 Meniere's disease patients. Thus far, 85 patients have been studied. Of these, nine had surgery and biopsies and had positive responses to glycerol (10 dB or greater threshold shifts at two low frequencies or a 10% improvement in their discrimination), no bony closure of the endolymphatic duct, and all of the other factors screened: hypothyroidism, glucose intolerance, vasomotor instability, allergic diathesis, and microcirculatory problems. Of these nine

patients operated, all had relief of their vertigo. In six the hearing has remained unchanged, and in three the hearing improved. The remaining patients are either undergoing further study or are being treated for specific problems discovered in their work-up: diabetes (2), hypothyroidism (2), perilymph fistula (1), basilar artery insufficiency and arteriosclerosis (21). Many of the patients had Meniere's syndrome (33) and not Meniere's disease, or the disease had progressed past the point where surgery would help.

Another investigator is studying the ototoxicity levels of ethacrynic acid and the combined effects of ethacrynic acid and known ototoxic antibiotics. Using electrophysiological techniques, ototoxicity of ethacrynic acid in different concentrations was established after intravenous injections. The greatest effects were observed in the hair cells of the inner ear. Decreases in the cochlear microphonics were noted, which remained for four hours with no tendency to recover.

A team of investigators has designed a probe tip assembly for use with electroacoustic impedance bridges which will fit a larger proportion of the clinical population than can be fitted presently with available assemblies. This group has also completed a pilot study of hearing of elderly people of various races in the Chicago area in an investigation of presbycusis. Although sample size is limited and no claims are made as to the representativeness of the sample or the validity of the aged population as a whole, many interesting findings were so clear that they strongly support the supposition that they would appear in a fully representative sample. One such finding is seen in the difference in hearing levels by race and sex. The mean air conduction pure-tone average for right ears in the sample of white males was 36.8 dB while for black males it was 30.6 dB; for white females 30.1 dB, and for black females 25.9 dB.

Inner Ear

In an investigation of the chemistry and physiology of inner ear fluids, one investigator determined that: (1) Human inner ear fluid samples (45%) are found to contain small amounts of erythrocytes after centrifugation. These samples also contained minute amounts of hemoglobin indicating that a certain amount of hemolysis had occurred. Most samples without erythrocytes did not have measurable amounts of hemoglobin. (2) The glucose concentration of the perilymph in acoustic neuroma and the endolymph in Meniere's disease resemble those found in the fluids of the normal anesthetized cat. (3) Sodium, potassium, glucose, and total protein concentration in uncontaminated endolymph samples from patients with Meniere's disease closely resemble endolymph from the normal anesthetized cat. This suggests that the defect in Meniere's disease is an abnormally large volume of endolymph with a normal endolymph chemical composition. (4) Preliminary experiments indicate that the cochlear microphonic recording does not significantly change after the blood sugar level decreases to 50 or 60 mg/100 ml in response to insulin given intravenously.

Another investigator is attempting to provide ultrastructural information on normal and pathological inner ears of animals, particularly to study the normal cochlear blood vessels and describe the structural alterations which

develop after interference with the main labyrinthine vascular supply or drainage site. In examining the blood vessels of the modiolus of the cochlea, he unexpectedly found fenestrated capillaries in the modiolar connective tissue which surrounds the major blood vessels. The only other place known to show fenestrated capillaries in the inner ear is the endolymphatic sac. In the modiolus, arterioles are different from those which are found in the membranous lateral wall and are of equal size; e.g., they show typical smooth muscle cells while the latter show more pericyte characteristics.

Another laboratory reported a new method of early diagnosis of acoustic neuroma. This disorder begins in the bony channel between the inner ear and the brain and is not malignant. The only known treatment is surgery for complete removal as early as possible to prevent pressure on the brain. The new procedure involves pasting four electrodes on the patient's face; one on each side of the nose and one at the outer end of each eyebrow. Using a weak electric current, the investigators stimulate nerves painlessly through the electrode on the nose to see whether or not the muscles controlled by the facial nerve respond normally. Next, they stimulate the eyebrow electrodes to study the effect on the supraborbital nerve. Photographs of the reactions are taken and displayed on an oscilloscope. The first five patients suspected of having acoustic neuroma all showed abnormal changes on one side of one or both nerves.

Cochlea

One investigator, in an attempt to elucidate the various stages of transduction and amplification of acoustic energy in the inner ear, studied the ear of the guinea pig. He found that the cochlear microphonics (CM) and the summating potentials (SP) induced by a wide range of frequencies and intensities of sound were recorded in the scala media in each of the four turns of the cochlea. A precise quantitative relationship was demonstrated between the cochlear potential and the parameters of the sound.

Another investigator sought to discover the projections of neurons in the cochlear nuclear complex of the cat in a quantitative study of the trapezoid body (TB). He found from degeneration studies that at the midline, fibers in the trapezoid body occupy specific zones and possess certain diameters depending on the locus of the lesion in the ventro-cochlear nucleus. An actual count of these axons in the normal preparation should shed light on the neuronal composition of the cochlear nucleus. A single two micron plastic section, cut perpendicular to the course of the trapezoid body near its decussation, was studied to determine the total number of fibers in the region and to determine the spectrum of fibers in different areas of the trapezoid body.

Comparative Hearing

In an investigation of coding in the avian auditory nerve, a group of investigators have successfully worked out recording and stimulation techniques and have recorded responses from more than 300 pigeon auditory nerve fibers. In order to compare results from birds with previous results from cats, they have also recorded from single fibers in the cat auditory nerve under conditions identical with those of the bird studies. All of the pigeon auditory nerve

fibers studied discharge spontaneously in the absence of any controlled stimulus. Rates of spontaneous activity range from about 10 spikes to over 200 spikes per second. The distribution of spontaneous rates for pigeons is significantly different from that in cats. Specifically, while no pigeon fibers have spontaneous rates of less than 10 per second, many cat fibers have rates of less than one per second. Conversely, many pigeon fibers discharge 150 per second, while few cat fibers are above 100 per second.

Another investigator, looking into the nature of genetic control of acoustical behavior in field crickets, measured behavioral response (singing in males and phonotaxis in females), electrophysiological monitoring of units involved in behavior, and genetic manipulation and interpretation of results obtained from behavior. This approach is aimed at understanding a relatively simple communication system in terms of genetic and developmental control, and to try to explain the evolution of many diverse signals used by singing orthoptera (crickets). A colony has been established and genetic hybrids were produced. They are ready to determine the extent that the female locomotes to the calling song of her own species and discriminates against other species.

Speech

A group of investigators is studying speech breathing mechanics in order to give a comprehensive account of the respiratory function in normal speech and disordered speech where respiratory dysfunction is involved so as to improve diagnosis and management of respiratory based speech pathologies. Emphasis is on understanding control mechanisms of speech breathing physiology at the neurological and mechanical levels, including a study of how forces provided by the respiratory pump in the form of pressures and flows act and interact with various structures of the head and neck to generate speech sounds. They have treated the chest wall as a two part kinematic system comprised of the rib cage and diaphram-abdomen in parallel, and wherein the volume displaced by each part is linearly related to the motions of the points within it. Using measurements of changes in anteroposterior diameters of the rib cage and abdomen, subjects were studied in upright and supine postures during several respiratory maneuvers and utterance tasks. Results were charted in relative motion diagrams (rib cage vs. abdomen) which included the relaxed configuration of the chest wall and departures therefrom during utterances. For conversation, reading, and singing, lung volume events were restricted to the mid-volume range and were dependent upon body posture and utterance loudness. Relative volume contributions of the two parts differed for subjects and utterances and ranged from all rib cage displacement to all abdominal displacement. During speech, the chest wall was distorted from its relaxed configuration and differently so in the two postures studied. Potential mechanisms responsible for these distortions lead them to conclude that the distortions observed constitute a "volume platform" or posturing of the chest wall, from off of which the speaker produces speech but does not further significantly distort the system in providing the changes in driving pressure required for typical utterances.

Another investigator has determined the acoustic characteristics of the speech of deaf children and how they differ from those of normal speech. The most common errors affecting intelligibility are of prime interest. These include errors involving individual phonemes as well as errors relating to

prosodic features. The speech of 40 deaf and 20 normal children was analyzed using the corpus of speech material obtained the preceding year. The acoustic speech signals were processed digitally to yield estimates of fundamental frequency, formant, and short-term energy contours for a selected number of utterances. The acoustic data correspond closely to the perceptual data. The measured fundamental frequency contours for children with pitch problems clearly reflected the perceptual judgements (pitch too high or too low or excessively variable). The normal children had average fundamental frequency values in the region of 200 Hz. The deaf children with abnormally high pitch had fundamental frequency values close to 300 Hz. For normal children, the fundamental frequency contour could be adequately approximated by a short-term orthogonal polynomial representation of degree no higher than a cubic polynomial over 80 msec. time intervals; for deaf children whose pitch was rated excessively variable, polynomials up to the fifth or sixth degree were required.

Similarly, articulatory errors are reflected in the structure of the formant and fundamental frequency contours. One of the most common articulatory errors is the omission of consonants. The corresponding acoustic analysis showed an almost complete absence of the expected formant transitions characterizing the intended consonant. In general, formant contours for the deaf speech appeared to be flatter and have less structure than those for normal speech. These findings provide, for the first time, a detailed quantitative description of both the perceptual and acoustic characteristics of the speech of deaf children and how these variables relate to various measurements of hearing impairment. From the data, development of experimental speech training aids for the hearing impaired will be forthcoming.

Vestibular System

An investigator utilized testing procedures based upon avoidance conditioning that determine the body equilibrium maintenance ability in squirrel monkeys. These testing procedures provide quantitative data about the functional alterations after ablation of different parts of the equilibrium system. A comparison can be made regarding the severity of dysequilibrium, the speed of functional compensation (or modification) and the maximum degree of functional recovery. Morphological confirmations, which include neurohistological investigations, are performed to validate functional data. The study of the correlation between bodily equilibrium function and oculomotor function after selected vestibular lesions has resulted in finding that after experimentally created cerebellar uvula and/or nodulus lesions in squirrel monkeys, the bodily equilibrium function, spontaneous nystagmus, positional nystagmus, postrotatory nystagmus, and optokinetic nystagmus were repeatedly observed for about three months. It was found that both areas are important for body equilibrium maintenance. However, the cerebellar nodulus was more important when compared to the uvula. As far as oculomotor function is concerned, the nodulus appeared to be facilitatory for optokinetic nystagmus and most probably acted as inhibitory for vestibular evoked eye movement.

Another investigator made an assessment of the role of the otolith organs in human vestibular dysfunction and developed a new test for assessing the integrity of these structures. He studied 12 patients in a new tilt-chair

device where the ocular countertorsion reflex is measured photographically by comparing eye torsion against a line drawn between the pupils of both eyes. This method requires no uncomfortable attachments to the subjects head and has resulted in the same degree of reflex activity and scatter of data between measurements as in the previous use of the head frame. He has studied this reflex in six patients with total inner-ear removal and confirmed earlier results of 50% reduction in reflex activity. He also studied five patients with postural vertigo where the reflex response was skewed in a direction not predicted by other studies, suggesting an irritative effect of the affected labyrinth.

OTHER SENSORY MECHANISMS

Olfaction

One group of researchers investigated the dependence of olfactory discrimination upon the interconnections of the bilateral structures of the olfactory system. After defining olfaction in animals with "split brains," tests were made to verify the role of olfaction for various structures assumed on anatomical grounds to be part of the olfactory system. Following callosalotomy, animals trained with a nasal plug took significantly longer to learn a two-odor discrimination than those trained without a plug. Since it was shown that a nasal plug in normal or sham operated animals did not affect learning rates, the deficit in callosalotomized plugged animals was attributed to the reduced ability to integrate the disparate messages of asymmetrical mucosal stimulation. On the average callosalotomized animals trained without a plug did take longer to learn but such effect was not statistically significant. Similarily callosalotomized animals, although demonstrating a greater deficit, did not significantly differ from sham operated animals following insertion or transfer of a nasal plug. These results are not consistent if callosalotomy affects integration of mucosal messages. The finding of qualitative, but not statistical consistency, suggests repetition with a new stereotaxic device.

Taste

In a comparative investigation of the sense of taste, a group of researchers demonstrated a need for zinc in humans for normal taste functions. They found however, that the taste preference behavior appeared unchanged in rats on zinc deficient diets.

This group also studied the effect of iron and protein deficient diets on taste behavior in animals. They found that protein deficient animals respond to taste stimuli as do calorically deprived animals. In studies on taste in hedgehogs, they determined that this animal differed from other mammals in terms of an indifference to common sugars.

Touch

An investigator is studying the parameters of cutaneous coding in order to determine the kinds of discriminations the skin can make when presented with mechanical stimuli, compare this to other senses and finally develop a cutaneous communication system. Subjects were tested with vibratory stimuli presented simultaneously to both the index and the little finger. The amplitude of

vibration required to be detected when a single site is being stimulated were compared to the amplitude of vibration required to be detected when both sites are stimulated simultaneously. Another experiment was to complete difference threshold (DL) measurements for low-intensity stimuli in the absence of masking vibration (quiet) and in the presence of three different intensity levels of a masking vibration. Results show that the general effect of the masking vibration is to raise the DL particularly at lower levels of the test stimulus.

Hunger and Thirst

A group of investigators studying the neurophysiology of food intake described the nature of the structures in the hypothalamic feeding centers that are concerned with the regulation of the intake of particular nutrients. The technique of injecting the toxic goldthioglucose (GTG) directly into the hypothalmus was combined with the cafeteria regimen. This permitted the measurement of voluntary protein and carbohydrate intake following a variety of lesions. A comparison in rats of electrically induced lesions and GTG lesions on cafeteria choice is underway to be followed by similar experiments involving monosodium glutamate and deoxyglucose injections. They have already demonstrated abnormalities of protein intake after MSG injections in mice. They have hypothesized that GTG combines irreversibly with the glucose receptors which appear to be situated on the surface of cells in the hypersensitive ventromedial nucleus. A detailed histological study of early degenerative changes in the hypothalamic neurons and small vessels following GTG is completed.

Another investigator completed a study on the antidromic and orthodromic activation of cells in the paraventricular nucleus on the control of oxytocin and secretion. It was possible to make a positive identification of neuroendocrine cells by showing antidromic activation of cells located in the paraventricular nucleus following stimulation via an electrode located in the hypothalamic hypophyseal tract. These cells were studied under three different conditions of anesthesia. In the first case, urethane anesthesia was used; second, an unanesthetized preparation was used; and in the third case an unanesthetized preparation with reserpine administration.

The proportion of cells antidromically activated in the paraventricular nucleus appeared independent of the anesthetic conditions. Similarly, the latency, the ability to follow high frequency stimulation of the hypophyseal tract was unaffected. The proportion of cells which showed spontaneous activity was in all cases similar and around 50%. In looking into the glucoreceptor mechanism of hunger, the investigator found that the infusion of 2-deoxy-D-glucose into the venous system of the rabbit produced increased eating within three hours following the beginning of the infusion. The most consistent finding, however, was that 2DG produced more eating over the three hour period with a shorter, more consistent latency, when infused into the portal system of the gut and liver (hepatic-portal system) than when injected into the jugular vein.

APPENDIX A

RESEARCH GRANTS AWARDED IN FY 1973 BY DISORDER CATEGORY

(Dollars in Thousands)

DISORDER CATEGORY	No.	AMOUNT	% of $
TOTAL ALL DISORDERS	1,056	62,414	100.0
1. NEUROLOGICAL DISORDERS			
A. Neurological Disorders of Early Life	158	8,120	13.0
B. Neurological Disorders of Aging	42	2,800	4.5
C. Cerebrovascular Disorders	56	6,400	10.3
D. Convulsive or Related Paroxysmal Disorders	33	2,885	4.6
E. Demyelinating or Sclerosing Disorders	41	2,400	3.8
F. Muscular or Neuromuscular	110	5,156	8.3
G. Infectious Diseases	10	540	0.9
H. Trauma or Injury	54	3,670	5.9
J. Tumors or Nervous System	18	898	1.4
M. Neuroendocrine Studies	70	2,800	4.5
N. Neural Aspects of Learning or Behavior	33	1,587	2.5
P. Nervous System Studies - Normal Function	126	5,500	8.8
TOTAL - NEUROLOGICAL DISORDERS	751	42,756	68.5

APPENDIX A

RESEARCH GRANTS AWARDED IN FY 1973 BY DISORDER CATEGORY (Contd.)

(Dollars in Thousands)

DISORDER CATEGORY		No.	AMOUNT	% of $
2.	SENSORY AND PERCEPTUAL DISORDERS			
	A. Disorders of Hearing or Equilibrium	117	7,676	12.3
	B. Disorders of Speech or Other Higher CNS Functions	31	1,840	2.9
	C. Disorders of Other Senses	132	5,010	8.1
	TOTAL - SENSORY & PERCEPTUAL DISORDERS	280	14,526	23.3
3.	MULTI-CATEGORICAL	17	5,020	8.0
4.	CONFERENCES	8	112	0.2

ANNUAL REPORT
July 1, 1972 through June 30, 1973
Extramural Programs
Training Grants and Awards Branch
National Institute of Neurological Diseases and Stroke

For four years the Institute's training programs have gradually been eroding since additional funds have not been provided to support training activities during inflationary years. Fiscal Year 1973 was to have been the year the Secretary, DHEW, was to take a major stand concerning training programs with the result that there would no longer be a "moritorium" on support for training activities. However, the President's proposed Fiscal Year 1974 budget reflected a decision to phase out Federal support for training programs.

In phasing out support for training, the guidelines drawn up by the NIH and the Office of Management and Budget specified that the highest priority for available funds be given to trainees to whom program directors had made firm commitments prior to January 29, the date the President submitted his Fiscal Year 1974 budget to Congress. Obviously, without any new trainees appointed subsequent to January 29, over a period of three or four years, the number of trainees receiving NINDS support would decrease and the phase out guidelines specified that support for the environment should be reduced proportionately.

Concerning the Institute's support for graduate training programs, the phase out guidelines were the same whether the program was in the last year of a project period or whether it had additional years of committed support. The major guideline was that beginning July 1, 1974 and thereafter, no new trainees could be supported with grant funds. Trainees currently in the program and new trainees who would be initiating their training programs prior to June 30, 1974 could be supported until their training was completed. Under these guidelines, a number of programs received essentially the same support beginning July 1, 1973 as they received July 1, 1972.

Concerning Postdoctoral Fellowships and Special Traineeships, the Institute proceeded through December as though there would be support for new applicants and a number of new awards were made. Teacher-Investigator Special Traineeship applications were reviewed at the January meetings of the training committees. Six applications were recommended for approval and the applicants were invited to Bethesda to be interviewed. However the interviews were cancelled and the applications were administratively withdrawn.

Concerning the Research Career Development Award program, the Institute was in the process of making eight new awards at the time of the January 29 Congressional message and we were allowed to fund those applications. All RCDA's will be honored through the total period of committed support.

Concerning Research Career Awards, it is the intent of the NIH that these awards be honored in accordance with the original guidelines, until the candidate retires or until he reaches his 70th birthday, which ever comes first.

For Fiscal Year 1973 the Department of Health, Education, and Welfare received no appropriation but instead the component parts of the Department supported ongoing programs through a series of "continuing resolutions."

To support the Institute's training grant and traineeship programs, $11.863 million were available. Broken down into programs, this provided $10.600 million for graduate training grants and $1.263 million for Special Traineeships. These funds enabled the Institute to support 201 training programs, 72 Special Trainees, and 13 Teacher-Investigator Special Trainees. (For the distribution of these awards among various training areas, see Appendices A, B, and C.) To support the Fellowship program, $1.919 million were available. These funds were used to continue support for 10 Research Career Awardees and to support 56 Research Career Development Awardees and 59 Postdoctoral Fellows. (For the distribution of these awards among the various training areas, see Appendix D.) For the scientific evaluation of applications, $.045 million were required.

According to the phase out guidelines, no new awards were made despite the fact that the National Advisory Neurological Diseases and Stroke Council recommended approval of 20 new applications. In addition, no approved supplemental applications or approved requests to expand programs were honored. All awards were made within the phase out guidelines and all were negotiated 10%, except for the "Centers of Excellence" which had committed funds only to support trainees.

BRANCH PERSONNEL

At the beginning of the year the professional staff of the Branch included the Chief and three Executive Secretaries: Dr. Harold Fournelle, Dr. J. Buckminster Ranney, and Dr. George Simon.

Since support for training programs is being phasedout, the two Neurology Committees were abolished and the Executive Secretaries of the Committees left the Institute. Dr. Fournelle retired at the end of February after 23 years of Federal employment, and Dr. Simon accepted a position with the Environmental Protection Agency beginning June 10, 1973.

Dr. Ranney is the Executive Secretary, Communicative Disorders Review Committee. The Committee was responsible for evaluating both training and program project applications in the communicative disorders areas. Since support for training programs is being phasedout, the Committee still functions to review program project applications. Dr. Ranney does not have responsibility for the administration of supported projects; the surveillance and administration of funded projects is the responsibility of the Research Grants Branch.

MANPOWER EVALUATION STUDIES

Four contracts, awarded to the American Neurological Association, the American Association of Neurological Surgeons, the American Council on Otolaryngology, and the American Speech and Hearing Association, to assess health manpower education and determine the training needs for the decade of the eignties, completed one year of work and applied for and were approved for six-month extensions to complete the contract objectives, including the preparation of final reports. To coordinate the activities of the four contractors, to assume uniformity in the data being collected, and to enable all contractors to benefit from unique approaches developed by any one contractor, Dr. David Fairbanks, as a consultant to the Institute, was involved in each study. Dr. Fairbanks terminated this relationship on May 14 and Dr. Raymond Summers, Chief, Training Grants and Awards Branch, assumed these responsibilities.

During the year the Institute recognized the need for a similar study covering the basic sciences of neurology and communicative disorders. Although a number of organizations specified an interest in a contract to study manpower needs in the basic sciences, in response to a "Request for Proposals," only two organizations responded, the National Academy of Sciences and the American Speech and Hearing Association. Both organizations submitted approvable protocols, but an Ad Hoc Technical Merit Review Committee gave the highest rating to the National Academy of Sciences' proposal. The Review Committee consisted of the following individuals:

Dr. Harry Doukas, Acting Chief,
Office of Research Manpower, DRG, NIH

Dr. Marvin Dunn, Associate Director,
Division of Physician and Health Professions
Education, BHME, NIH

Mrs. Elizabeth C. Hartman, Director of Operations,
Joint Committee for Stroke Facilities

APPENDIX A

Distribution by Scientific Fields,
of Training Grants Awarded in FY 1973

FIELD	NUMBER	AMOUNT
Audiology	6	$ 272,300
Cerebrovascular	1	86,500
Child Neurology	15	782,100
Communicative Disorders	6	411,400
Neuroanatomy	3	139,500
Neurobiology	4	160,400
Neurochemistry	2	90,900
Neurological Sciences	6	194,400
Neurology	54	3,456,900
Neuropathology	13	501,000
Neuropharmacology	2	128,900
Neurophysiology	13	605,800
Neuroradiology	9	359,300
Neurosurgery	23	921,600
Neurovirology	1	36,500
Otolaryngology	38	2,267,600
Sensory Physiology	3	126,700
Speech Pathology	2	58,200
TOTAL	201	$10,600,000

May 1973

APPENDIX B

Distribution, by Scientific Fields,
of Special Traineeships Awarded in FY 1973

FIELD	NUMBER	AMOUNT
Audiology	1	$ 15,300
Biochemistry	2	28,500
Biophysics	2	26,800
Child Neurology	15	192,400
Immunology	2	29,300
Neuroanatomy	1	9,500
Neurobiology	4	46,700
Neurology	2	20,300
Neuropathology	5	79,400
Neuropharmacology	4	57,500
Neurophysiology	10	130,400
Neuroradiology	15	216,800
Neurosurgery	5	83,200
Neurovirology	2	31,500
Sensory Physiology	2	17,200
TOTAL	72	$ 984,800

May 1973

APPENDIX C

Distribution, by Scientific Fields,
of Teacher-Investigator Awards Granted in FY 1973

FIELD	NUMBER	AMOUNT
Cerebrovascular	2	$ 44,300
Immunology	1	19,500
Neuroanatomy	1	22,500
Neurobiology	1	19,500
Neuroendocrinology	1	22,500
Neurology	1	22,500
Neuropathology	1	18,200
Neurophysiology	2	43,000
Neurosurgery	1	23,500
Neurovirology	1	23,500
Sensory Physiology	1	19,400
TOTAL	13	$ 278,400

May 1973

APPENDIX D

Distribution, by Scientific Fields, of Fellowships Awarded in FY 1973

Field	RESEARCH CAREER AWARDS No.	RESEARCH CAREER AWARDS Amount	RESEARCH CAREER DEVELOPMENT AWARDS No.	RESEARCH CAREER DEVELOPMENT AWARDS Amount	POSTDOCTORAL FELLOWSHIPS No.	POSTDOCTORAL FELLOWSHIPS Amount
Audiology	1	$ 29,700	2	$ 29,800	2	$ 18,500
Biochemistry			3	51,900	6	61,200
Biophysics					1	11,100
Cerebrovascular	1	30,400				
Child Neurology					1	16,500
Communicative Disorders			1	20,300	1	6,500
Epidemiology			1	32,700		
Immunology			1	22,000		
Neuroanatomy			3	65,200	8	63,800
Neurobiology			6	109,500	5	43,400
Neurochemistry			8	165,600	8	57,900
Neuroendocrinology	1	25,400	2	45,600	2	15,500
Neurology	2	61,700	1	25,000	1	7,000
Neuropathology			2	46,800	3	31,800
Neuropharmacology	2	60,100	1	19,000	2	14,500
Neurophysiology	1	7,600	15	301,300	15	132,600
Neuroradiology					1	14,900
Neurosurgery			1	21,000		
Neurovirology			5	100,900		
Otolaryngology	1	33,000				
Physiological Psychology	1	31,300	1	16,000	1	9,500
Sensory Physiology			2	43,700		
Speech Pathology					2	15,000
TOTAL	10	$ 279,200	56	$1,116,300	59	$519,900

May 1973

DATE DUE

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